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THE VARIABILITY OF LOAF VOLUME IN EXPERIMENTAL BAKING

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When efforts to unify baking procedure were first being brought before the American Association of Cereal Chemists, Dunlap (1926) drew attention to the importance of the personal factor in its effect upon the volume of loaves baked from the same flour by several workers under conditions that were as nearly identical as it seemed humanly possible to make them. He concluded from his unpublished data that the manipulation of doughs manually in a uniform manner by different workers presented a problem of great importance in the standardization of baking procedures. In the same year Blish (1926) stated, "No two cereal chemists working under identical conditions . . . will be likely to bake identical loaves from the same flour. . . . The personal touch of the operator in knocking down and molding the dough is the principal cause of . . . variations." Blish also urged that the personal equation must be reduced to the minimum, and expressed the opinion that it could only be achieved by means of mechanical devices to replace the use of hands where a sense of touch was involved.

Largely through the efforts of Blish and Harrel, working at the head of special committees dealing with the experimental baking test, the Association has accepted a "basic standard procedure" of experimental baking (Blish, 1928a) and considerable progress has been made in surmounting the many difficulties presented. However, little advance has been made in dealing with the problem indicated by Dunlap. Moen (1929), at the request of the baking committee of that year, gave special attention to this phase of the work and found that, following explicit directions as to number of turns and folds, "two experienced operators molding apparently in much the same way consistently produced different results on

the same flour in respect to volume," and two other experienced workers "who apparently exhibited considerable difference in manner of molding, produced similar results in volume and internal characteristics." As a result of the committee's extensive work in 1928-9, Harrel (1929), impressed by distinct "panning personalities" shown by different collaborators, again stressed the opinion that variation in loaf volume is probably largely due to the manner of molding the loaf. Harrel, at that time, was led to state: "As scientists, let us not dismiss lightly the lack of agreement in our results. A tentative method has been adopted, but to perfect it will require several years of intensive research on factors causing these variations."

The present study represents part of a first effort to indicate the nature, and possibly the sources, of variation in volume of replicate bakes of bread from the same flours. The results have seemed of sufficient importance in view of current interest and endeavor in experimental baking to warrant a preliminary communication.

The variability of an end quantity is a resultant of the variabilities of the individual procedures and materials involved in obtaining that final result. The cause of variability of the end result cannot be said to be known until that variability is measured and is shown to be equal to the given function of the minor variabilities involved. It is important to recognize this, particularly as correlation between end results is materially reduced as the extent of minor variabilities increases. For example, the correlation between protein content of flour and loaf volume is undoubtedly considerably reduced by the variability of personal judgment in protein determinations and personal touch variations in dough handling. It becomes of prime importance to arrive at a valid measure of the significance of these minor variations, which, from a relative point of view, may be of major importance.

It is somewhat surprising that prior contributions have not been made toward measuring the variability to be expected at the hands of any worker in making replicate bakes of the same flour. A survey of the literature has failed to give satisfactory indication that this variability is known with precision in any published case, nor do the authors believe that the extent of the variability which may be expected is adequately appreciated.

Experimental

All of the baking work considered herein was carried out in

the laboratories of the Department of Chemistry, University of Saskatchewan, by men of considerable experience in this work. The routine of the investigations at that laboratory demands a baking capacity of the operators of up to 50 loaves per day, or more in exceptional circumstances. The technique of baking followed represents as close an approximation to that called for by the "basic standard method" as has been found compatible with the demands of the work. Doughs were not mixed by hand but with a Hobart mixer, with two minutes on first speed, one on second and a "clean up" of up to one minute on third. Molding was performed on a shielded and warmed board bench with the least possible use of dusting flour to facilitate speed of work. Loaf volume and weight determinations were made 20 minutes after the loaves came from the oven. In a few instances individual loaves were rejected where a known factor or factors came into play that might cause differentiation of that loaf or loaves from strict conformity with the other members of the series. Thus a drop of two degrees for a few minutes in the temperature of the proofing cabinet led to the rejection of six loaves likely to be affected, although but three of those loaves showed any influence of the change.

Three different bakers, designated in the following discussion as A, B and C, took part in the work. A was the most experienced, but at the commencement of work had not baked intensively for several months. It does not seem possible from the results to demonstrate that this factor had any appreciable effect. B and C were fully in the swing of daily baking routine. 532 loaves of bread were baked in ten working days, 326 of which were from a standard bleached flour (Standard 2) of the baking quality of which six months experience was available. Standard 3 was a fairly new flour (unbleached), which had only been tested for a few weeks. Both of the above flours were commercially milled from Canadian Hard Red Spring wheat and were carefully chosen as being desirable standards of comparison for the experimental baking work. Another flour was synthesized from remnants of low quality experimentally milled flours from Hard Red Spring wheats as a contrast to the two standards. It was very carefully mixed to insure uniformity and was baked in only one test.

The pans used were of two types. Those designated hereinafter as "tall" are the regular pans called for by the standard method of experimental baking. They are not the style of pan favored by the cereal research laboratories of western Canada and were used solely for comparison in two series where they were

alternated with the pans regularly employed. The latter are designated as "squat" pans, and have the following dimensions:

Top: 10 x 6.3 cm.
Bottom: 7.5 x 4.3 cm.
Depth: 4 cm.

Details of the series of bakes are summarized in Table I.

TABLE I

SUMMARY OF THE TEN SERIES OF BAKING TESTS CONSIDERED

Bake Series No.	Flour	Quantity	Modification of Basic Method	Baker	Pans Used	Number of Loaves
1	Standard 2	100 g.	none	A	squat	50
2	Standard 2	100 g.	none	B	squat	50
3	Standard 2	100 g.	none	C	squat	50
4	Standard 2	100 g.	none	A, B, C	squat	72
5	Standard 2	100 g.	none	A	squat	50
6	Standard 2	100 g.	none	A	squat and tall alternately	54
7	Low Quality	100 g.	none	A	squat and tall alternately	56
8	Standard 3	100 g.	none	A	squat	50
9	Standard 3	100 g.	no sugar	A	squat	54
10	Standard 3	100 g.	plus bromate no sugar	A	squat	46

Results

a. **Frequency Distribution Studies**—Detailed tables of the 532 individual volumes, with weights and temperatures where taken, will not be given in that form since it is possible to present all the salient features of the study in tables of constants and a few typical graphs.

As the first undertaking of this study, A, B and C on successive days baked 50 loaves each from flour Standard 2 in order that the distribution of volumes for each baker might be measured and any significant differences between the results of the three operators revealed. The constants for these series are presented in Table

TABLE II

FREQUENCY CONSTANTS FOR THREE OPERATORS BAKING 50 LOAVES EACH FROM THE SAME FLOUR (STANDARD 2)

Frequency Constant	Baker A	Baker B	Baker C
Mean	627.5 ± 1.4 cc.	624.0 ± 1.1 cc.	617.5 ± 1.0 cc.
Standard Deviation	14.6 ± 1.0 cc.	11.4 ± 0.8 cc.	10.1 ± 0.7 cc.
Actual Range	57 cc.	45 cc.	45 cc.
Probable Error of One Determination	9.8 cc.	7.7 cc.	6.8 cc.
Theoretical Range of 99% of Observations	75.1 cc.	58.7 cc.	51.8 cc.

II. To indicate the type of the distribution for loaf volume, the histograms of this variable are reproduced in Figure 1.

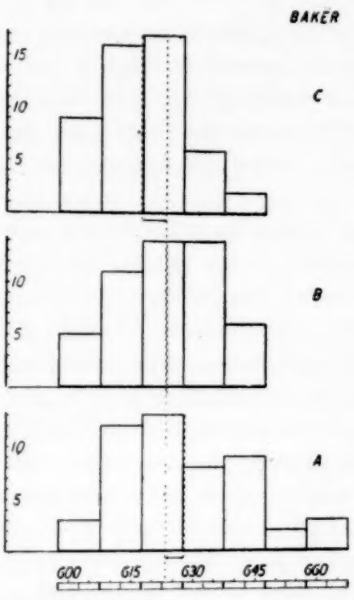

Fig. 1. Histograms of the distribution of loaf volumes obtained by three workers when baking fifty replicates from the same flour.

Averages are given by the dotted vertical lines. The classes of loaf volume have been given a range of 10 cc. to obtain adequate smoothing.

Two facts revealed by this comparison are striking. First, the three bakers, trained in the same laboratory, obtain very significantly different results in the average volumes secured. Furthermore, A shows considerably greater variability than B or C. Second, the actual total range of volumes for fifty loaves varies from 45 cc. to 57 cc. for the three workers. In view of the total spread of the loaf volumes in each case, the differences between the averages obtained by the three workers may appear to be of small importance. They are, however, very significant when compared to their probable errors.

The graduation of these histograms by normal curves would not seem justifiable in individual cases. That this is in all probability due to the small number of replicates in each series is illustrated by Figure 2, where the normal curve is super-

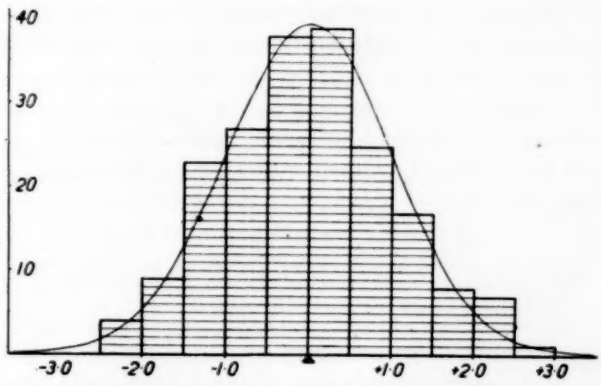

Fig. 2. Histogram and fitted normal curve of the combined distributions for loaf volume obtained by A in bake series Nos. 1, 5, 8 and 9. Grouping is in terms of $\frac{(x-\bar{x})}{\sigma_x}$ to eliminate disorderly differentiation of the four series.

imposed on the histogram of the sum of four simple replicate series of baker A (bake series Nos. 1, 5, 8 and 9), which have been grouped together in terms of relative deviations from the mean (or $\frac{x-\bar{x}}{\sigma_s}$) to remove the influence of disorderly differentiation between them. The graduation provided by the normal curve is highly satisfactory. Applying Pearson's (1900) χ^2, P criterion we find that the odds that the lack of exact agreement between the histogram and the normal curve is due to chance factors alone are approximately 3 to 1 in favor of concordance.

If, then, we may accept the normal curve as providing a suitable smoothing of the frequency distributions for the three bakes if a much larger number of replicates were available, an infinitely large series should show ranges for the three bakers of 75.1, 58.7 and 51.8 cc. for the central 99% of the distribution. This means that baker A, in comparing the *average* volumes of duplicate bakings of two different flours, would need to find a difference of at least 30 cc. in those averages before he could be justified in concluding that his two flours were different in their capacity to bake into large loaves of bread. The difference for baker C would need to be at least 20 cc. For single bakes these differences would have to be magnified by the square-root of two, i.e., they would become 40 cc. for A, 28 cc. for C. Baker B is intermediate in variability between A and C. The foregoing differences represent chances merely of twenty to one against the two flours being undifferentiated in the capacity considered.

In order to test the significance of molding and panning personality of the bakers in its influence on these different averages secured for the three workers, A mixed 72 doughs from the same flour (Standard 2) at intervals of five minutes and attended to all details of dough handling during the fermentation period. Then A, B and C molded and panned the doughs in rotation and the resultant volumes of the 24 loaves for each operator were measured as before. The constants of the frequency distributions are given in Table III.

TABLE III

FREQUENCY CONSTANTS FOR LOAF VOLUMES OBTAINED BY THREE OPERATORS WHEN MOLDING AND PANNING DOUGHS PREPARED BY ONE OPERATOR ONLY (FLOUR STANDARD 2)

Frequency Constant	Baker		
	A	B	C
Average	625.2 cc.	609.0 cc.	616.3 cc.
Standard Deviation	13.0 cc.	12.3 cc.	16.7 cc.
Actual Range	69 cc.	55 cc.	65 cc.

During this experiment it was noted that B was molding more slowly than usual and was exercising pains in handling that might have an effect upon the results. C seemed to be working in his usual manner. These observations have proven to be of value in explaining the results, for A and C have produced loaves of average volume very similar to those of their previous independent series, while B's average fell from 624 cc. to 609 cc. C showed a pronounced increase in variability of his results. This may be a reflection of the unusual circumstances of the work, as was more directly observable in the case of B. Both B and C were molding at 15 minute periods instead of the usual five minutes, and had no matters of mixing or punching to attend to in the interval. A was working very nearly under usual conditions.

While it may not advisedly be stated from this limited work that the different results obtained by these operators were due entirely to "molding and panning personality," it is quite obvious that the latter may well have been the major factor in causing the differentiation between these results. To what extent a mechanical molder would have reduced the variability of the results is not certain. The three workers may not have shown any differentiation in their average results if a mechanical molder had been available for the first three series of bakes.[1]

As the demands of other investigations necessitated that any further baking studies on this project be done by baker A alone, work on the personal factor was discontinued and attention given to certain other variables which seemed readily controllable.

In bake series No. 5, doughs from 50 g. quantities of flour (Standard 2) were mixed and baked in "squat" pans of proportionally smaller dimensions in order to see what change would ensue in the relative variability of the volumes.

The frequency constants

$$\text{Average} = 297.0 \pm 0.8 \text{ cc.}$$
$$\text{Standard deviation} = 8.5 \pm 0.6 \text{ cc.}$$

indicate a considerable decrease in absolute variability when the smaller amount of flour is used. If the standard deviations be considered in relation to the size of loaf, as given by the following coefficients of variation:

$$\text{100 g. of flour (Bake No. 1) C. V.} = 2.33\% \pm .16\%$$
$$\text{50 g. of flour (Bake No. 5) C. V.} = 2.85\% \pm .19\%$$
$$\text{Difference} = 0.52\% \pm 0.25\%$$

it will be seen that the difference in relative variability is only twice its probable error and hence it may not be considered significant.

[1] Since the preparation of this manuscript Fifield and Weaver (1930) have demonstrated a very pronounced reduction in the variability of loaf volumes when machine molding was substituted for the manual procedure.

Lewis and Whitcomb (1928) have made the most complete study available of the influence of size and shape of pan on the volume of the loaves of bread baked in them. Their investigation did not embrace pans for loaves made from 100 g. portions of flour, although it is possible that analogies would apply. It was thought that it might prove of value in the present study to compare the frequency constants when both the squat and the tall pans were employed. Accordingly these pans were used alternately in two series of bakes. The flour employed for the first series was again Standard 2, while the second series was baked from the specially prepared mixture of remnants of low quality flours. The constants derived from these series are presented in Table IV.

TABLE IV

FREQUENCY CONSTANTS SHOWING THE INFLUENCE OF TWO DIFFERENT TYPES OF PAN ON LOAF VOLUME FROM TWO FLOURS

Frequency Constant	Flour			
	Standard No. 2		Low Quality	
	squat pan	tall pan	squat pan	tall pan
Average	621.3±2.1 cc.	557.2±1.9 cc.	441.6±1.3 cc.	421.4±1.5 cc.
Difference (D)	64.1±3.0 cc.		20.2±1.8 cc.	
D/P.E.$_D$	21.2		10.9	
Standard Deviation	15.8±1.5 cc.	14.9±1.4 cc.	9.8±0.9 cc.	11.7±1.1 cc.
Difference	0.9±2.0 cc.		1.9±1.4 cc.	

They show that in the squat pans the doughs attain a significantly larger final volume. It is of interest to note that the increase in volume is three times as great with the standard flour as with the low quality flour. The variabilities of the loaf volumes from the two types of pan do not appear to differ consistently or significantly, but the variation is higher in both cases for the standard flour than for the low quality mixture. Differences in texture seemed to be more imaginary than otherwise, except for the shape of the vesicles. The latter were more rounded for the squat pan loaves.

A comparison of loaf weights was also made with these two

TABLE V

FREQUENCY CONSTANTS SHOWING THE INFLUENCE OF TWO DIFFERENT STYLES OF PAN ON THE WEIGHT OF LOAVES FROM TWO FLOURS

Frequency Constant	Flour			
	Standard No. 2		Low Grade	
	squat pan	tall pan	squat pan	tall pan
Average	144.3±.2 g.	142.3±.3 g.	146.2±.21 g.	144.5±.22 g.
Difference D	2.00±.33 g.		1.75±.32 g.	
D/P.E.$_D$	6.0		5.46	
Standard Deviation	1.65±.15 g.	2.27±.21 g.	1.97±.18 g.	1.76±.16 g
Difference	−0.62±0.23 g.		+0.21±0.24 g.	

series baked in different pans. The resulting constants, given in Table V, indicate a significantly lower weight of about 2 g. in the loaves baked in the tall narrow pans called for by the basic method. The standard deviations in the two cases are not significantly different.

The work was continued with the new flour (Standard 3) which was being introduced at that time as laboratory standard. Fifty loaves were baked from this flour by the basic formula and then a series of 54 doughs were mixed without sugar and baked, followed next day by another series of 46 doughs having no sugar but with 1 cc. of a 0.1% $KBrO_3$ solution added. The bromate differential test (originated by Werner) has received official commendation by Blish (1928b) as supplementary test "C" to the basic standard formula, and its high importance in revealing the baking quality of Saskatchewan wheats has more recently been extensively determined by Larmour (1930).

The results of this study of the effect of omitting sugar from and adding bromate to the dough are in part shown by the frequency distribution constants presented in Table VI.

TABLE VI

FREQUENCY CONSTANTS SHOWING THE EFFECT ON LOAF VOLUME OF CERTAIN MODIFICATIONS OF THE BASIC FORMULA

Frequency Constant	Modification of Basic Procedure		
	None	No Sugar	Plus Bromate No Sugar
Number of loaves	50	54	46
Average	623.0±1.1 cc.	515.2±2.1 cc.	605.8±1.9 cc.
Standard Deviation	11.3±0.8 cc.	23.4±1.5 cc.	18.9±1.3 cc.
Actual Range	50 cc.	90 cc.	75 cc.
Theoretical Range of 99%	58 cc.	120 cc.	97 cc.

It is worthy of note that this standard flour gives a significantly lower average volume than does Standard 2 at the hands of baker A. The difference of 4.5 cc. on the average is, however, small. More important is the fact that A has in the above series secured decidedly less variability in the loaf volumes from this standard flour than he obtained from Standard 2. Whether there is a difference in the sensitivity of the fermented doughs or whether A exercised a more uniform molding technique this day is beyond answering here.

Loaves of fair volume (515 cc.) result when sugar is left out of the formula with this flour. However, it represents a decrease of 92 cc. from the average obtained with the inclusion of sugar.

Far more striking is the increase in variability of loaf volume to double its previous magnitude. This variability is reduced and the average volume increased by the introduction of 0.001% $KBrO_3$ into the formula. However, the addition of the potassium bromate does not fully compensate for the omission of sugar. The recovery in average volume represents 84% of the fall occasioned by omitting sugar. The decrease in the standard deviation is not quite half of the first increase. The average weight of the loaves when bromate was used was $2.01 \pm .19$ g. less than those from the sugar-free dough alone. The drop is decidedly significant and certainly indicates either a modified substratum conducive to more rapid fermentation by the yeast, or perhaps that a stimulus is given to the enzymatic activity of the organism by the addition of the bromate.

b. **Correlation Studies.**—This investigation has provided data for considering the interrelationship between certain variables. Attention may be first given to the effect of the temperature of the dough as it leaves the mixing machine on the volume and the weight of the resulting loaf. If there be linear relationships between the initial temperature of the dough and these measures of the loaf, then a correlation coefficient between the absolute values involved should reveal them. If, however, there is an optimum initial temperature of the dough of 30° C. to which fermentation is sufficiently sensitive that departures of one or two degrees either way will affect the loaf volume or loaf weight, then correlations between the latter two variables and deviation without regard to sign of the initial temperature from 30° C. should show significant positive or negative values. In the laboratory work efforts were made to keep the initial temperatures of the doughs between 29° C. and 31° C. They rarely deviated more than 2° from 30° C.

The correlations of loaf volume and loaf weight with temperature have been computed where the needed data was obtained. The coefficients are presented in Table VII. Care has been taken to avoid spurious values of the coefficients due to disorderly differentiation by treating each sub-series separately and combining the correlations with proportionate weighing into an average for each type. From the results obtained it is clear that if the initial temperature of the dough has had any influence on the volume or weight of the loaves baked in these series, it is completely masked by the variability secured in the normal routine of replicate baking.

That is, until the latter variability is reduced considerably below its present magnitude, deviations of up to 2° C. of the initial temperature of the small dough from 30° C. may be tolerated in experimental baking without concern. The statement of Harrel (1926) that temperature of the dough after mixing bears a very important relationship to loaf volume is unfortunately not supported by accompanying data, and so we are precluded from considering the above experience in relation to his findings. Harrel's conclusions probably refer to doughs from 350 g. samples of flour. The importance of deviation of temperature from 30° C. after mixing is doubtlessly related to the size of the dough (assuming an otherwise constant baking technique). Attainment of temperature equilibrium in the fermentation cabinet may possibly be quite rapid for doughs from 100 g. of flour.

TABLE VII

CORRELATION BETWEEN INITIAL TEMPERATURE OF THE DOUGH AND THE VOLUME AND WEIGHT OF THE RESULTING LOAF

Bake Series No. and Subseries	Loaf weight and temperature of dough	Loaf weight and deviation of temp. from 30°	Loaf volume and temperature of dough	Loaf volume and deviation of temp. from 30°
1	+.05 ± .10	−.24 ± .09		
2	+.01 ± .10	+.09 ± .09		
3	+.04 ± .10	−.05 ± .10		
4 Baker A	+.28 ± .13	−.09 ± .14	+.02 ± .14	−.27 ± .13
4 Baker B	+.25 ± .13	−.07 ± .14	+.23 ± .13	+.03 ± .14
4 Baker C	+.43 ± .11	+.21 ± .13	−.18 ± .13	−.08 ± .14
6 squat pans	−.27 ± .12	+.20 ± .12	−.10 ± .13	−.05 ± .13
6 tall pans	+.42 ± .11	+.28 ± .12	−.15 ± .13	−.19 ± .13
7 squat pans	−.16 ± .12	−.17 ± .12	−.01 ± .13	−.20 ± .12
7 tall pans	+.16 ± .12	−.23 ± .12	+.04 ± .13	−.10 ± .13
Average	+.09 ± .04	−.02 ± .04	−.02 ± .05	−.12 ± .05

It was suggested by one of the bakers taking part in this work that the fermentation cabinet might not be uniform throughout in heat and draft characteristics despite the very careful endeavors to overcome such undesirable features by building in suitably placed baffle plates and ventilator slots. If place differentiation existed in the cabinet and was of such magnitude as to affect the doughs in the fermentation jars, then a positive correlation should exist between loaves baked from doughs which occupied the same position in the cabinet in different bake series. To test this point correlation coefficients have been calculated between the possible combinations of the first three bake series. These coefficients, submitted in Table VIII indicate clearly that cabinet position of the dough has had no sensible influence on loaf volume in these experiments.

TABLE VIII

CORRELATIONS BETWEEN LOAF VOLUMES FOR CORRESPONDING DOUGH POSITIONS IN THE FERMENTING CABINET

Paired Series	*N*	Coefficients of Correlation
1 and 2	48	$+.20 \pm .09$
1 and 3	48	$+.02 \pm .10$
2 and 3	50	$-.12 \pm .09$
Average	146	$+.03 \pm .06$

With the initial temperature of the dough after mixing and any unknown characteristics of the cabinet eliminated as factors of any major importance directly causing or contributing to the variability found among replicate bakes in this study, attention may now be directed again to the operators themselves. Do the replicate loaf volumes arising from the work of any one baker vary in a purely random manner about a constant average, or does increasing fatigue or changing disposition of the worker cause a systematic change in level of the average loaf volume during the period of the day's work? If the latter be true, then the variability of replicate bakes must be in part a function of the time at which they are made. To test for systematic variation one may measure the correlation between the volume of each loaf and that of its succeeding member in the series comprising the work of the period of replicate bakings. These correlations have been computed in all cases where the day's work involved an unbroken series of replicates, and are given in Table IX.

TABLE IX

CORRELATIONS BETWEEN LOAF VOLUMES IN A SEQUENCE IN TIME (BAKE SERIES NUMBER GIVEN IN PARENTHESES)

Baker A			Baker B			Baker C		
	N	*r*		*N*	*r*		*N*	*r*
(1)	49	$+.23 \pm .09$	(2)	49	$+.17 \pm .09$	(3)	49	$-.03 \pm .09$
(5)	49	$+.13 \pm .09$						
(8)	49	$+.11 \pm .10$						
(9)	53	$+.60 \pm .06$						
Weighted Average		$+.27 \pm .04$						

The data for two of the workers is too scant for satisfactory conclusions, but from the results of baker A there is clear evidence that his loaves have tended to vary in average volume systematically throughout the day. This tendency, as measured in one case by a correlation of $+.60 \pm .06$, may become of considerable importance. The standard deviation of loaf volumes at any instant of time in this series becomes 18.8 cc. as compared with 23.4 cc. for the

whole day. While this particular case contributes considerably toward the highly significant average value of r ($+.27 \pm .04$) for Baker A, the average value of the remaining coefficients ($+.16 \pm .05$) is also significant. The suggestion that such systematic variation in loaf volume may reflect yeast activity in some way must also be borne in mind until the contrary is proven. More extensive work on this phase is entirely desirable and indeed will be necessary before a fixed tendency to show secular variation is proven.

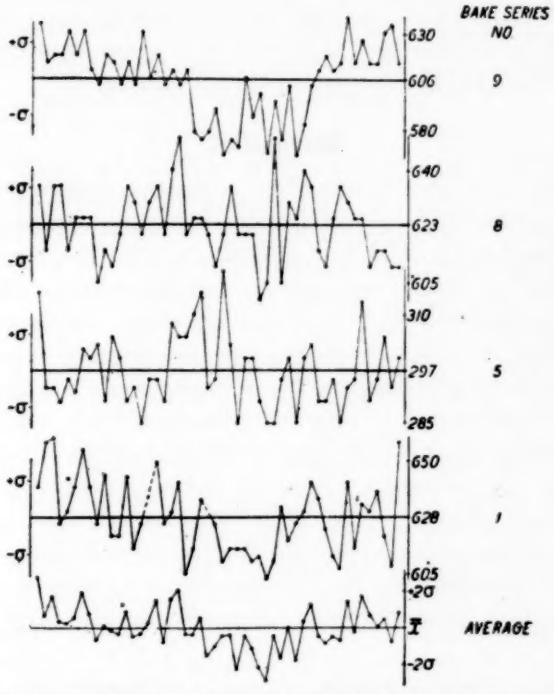

Fig. 3. Chart of the distribution of consecutive loaf volumes about the average for the series secured by the same baker in four series of replicate bakes. Deviations are in terms of the same scale of $\frac{(x-\bar{x})}{\sigma_x}$ for each series, the first bake of the day being on the left in each case. The mean deviations are given by the circles in the lower panel corresponding to the four series of solid dots above.

In order to study the nature of the systematic variation in the volumes obtained by baker A, the deviation of each loaf from the average of its series has been obtained and expressed in terms of the standard deviation of the series. By this means it has been possible to group the corresponding loaves (in point of view of time) in the four series and calculate the mean deviation from the general average. The results for each series and for the average

have been charted and are presented as Figure 3. The average for each series is given by the horizontal line and the consecutive loaf volumes are represented by dots (or circles for the mean deviations) joined by the lighter lines. As also measured by the coefficients in Table IX, the systematic variation is shown to be most pronounced in bake series Nos. 9 and 1. The tendency in both these series is very clear. The baker produces high loaf volumes at the beginning of the series, the volumes falling gradually to well below the general average at one-half to two-thirds through the sequence, then rising slowly to finish above the general average. It is apparent that this tendency is not manifest on all days, but it is the only clear one and is sufficiently pronounced to carry the mean deviations along the same general course.

Discussion

There is little doubt but that scientific workers engaged in experimental baking have an impression that the volume of the loaves of bread they bake are subject to a fairly large (or small) "experimental" error. Duplicate loaves sometimes agree in volume; they more often do not. If the difference between the duplicate loaves be of a much larger magnitude than is customarily encountered, or larger than the discrepancy usually tolerated by the worker, the bake is not infrequently discarded and another pair of loaves prepared, or, a third loaf is baked and the average of the two in closest agreement, or in some cases of all three, is accepted as the measure desired. The elimination from the average of a divergent member of a trio of bakes on account of unknown factors supposedly differentiating it from the other two is, in view of the foregoing results, probably quite unjustifiable in a large proportion of cases.

The magnitude of variation in loaf volumes from experimental replicate bakings demonstrated in this study is such that careful consideration must be given to the degree of confidence warranted by an average of duplicate bakings. That the volumes of loaves from a fine quality flour prepared and baked under supposedly standard conditions varied over an actual range of as much as 57 cc. in 50 replicates in this investigation, and might reasonably have been expected in a very large series to have varied over as wide a range as 75 cc. in that particular case, indicates that duplicates may be expected to give averages varying over a range of up to 50 cc. While this variation is that of workers in one laboratory only, it is quite possibly representative of a larger number of ex-

perimental bakers of high repute. That variation of the temperatures of the doughs immediately after mixing within a range of 30° C. ± 2° has not had any appreciable influence on loaf volume in this experimental baking is surprising in view of the importance of rigidly controlled mixing temperatures in commercial production.

Manual molding of the doughs appears in the present study to be a factor of very great importance in differentiating bakers. It is an entirely logical conclusion that the same factor may be largely responsible for the variation of replicates from the hands of any one operator. The latter thought is given weight by the foregoing demonstration that a particular baker showed a decided tendency to vary systematically throughout a day's work. The theory may not be tested adequately, however, until mechanical molders for the small experimental doughs are available and are so constructed that they compare favorably in operation with those of the commercial scale. When such apparatus is available, more rigid temperature control in fermentation and proofing may prove to be distinctly more important than has been demonstrated herein. At present, at least, experimental baking is subject to factors producing variation in loaf volumes that are undoubtedly of greater importance than they have been estimated to be by workers in this field.

The average discrepancy between duplicates undoubtedly has been conjured in many cases to represent the range within which loaf volumes are "accurate." The literature contains only very scant data that may serve to give one an idea of the variability of replicates or the range of duplicates as usually only the average is published. Moen (1929), however, gives some volumes of triplicate bakes with the basic formula that are of particular interest in connection with the present work. In Table I of his report, the four triplicates vary over ranges of 25, 25, 10 and 25 cc., the most divergent one in each of the 25 cc. ranges being omitted by him from the average. In Table II, also, the ranges vary from 15 to 60 cc., while in Table III the range is as high as 75 cc. in a triplicate bake. Worker A in Table III baked 12 replicate loaves varying from 450 to 535 cc. (latter eliminated) thus obtaining results comparable to, or greater in degree of variation than that obtained by the workers in the present paper. In striking contrast is the work of Herman and Hart (1927) who published duplicate tests wherein the loaf volumes of a pair were in perfect agreement in 41 cases out of 107, and only differed by 5 cc. in the remaining

66 cases. This surely would represent delightful accuracy in baking technique, and would be a most disturbing standard to all other workers were it not for the fact that the authors state all tests were made in triplicate, "while, *for convenience,* only duplicate loaves were tabulated" (the italics are ours). As the published pairs differ *solely* in volume, closest agreement in that characteristic seems to have been the basis of selection of the pairs. Such action, if taken and not explained in the paper, can only result in a damaging false confidence. Such agreement, if the pairs could possibly represent random selection, would seem to justify the measurement of volumes in intervals of a single cubic centimeter instead of the 5 cc. intervals employed.

The writers do not in any sense contend that the data of this study represent standards for other laboratories. Indeed a prominent feature of the work is the importance of the personal sense of touch. However, it is manifest that experimental laboratories must individually pay close attention to the variability encountered in replicate bakings before establishing criteria of the significance of the differences they obtain between different flours. It is in large part with the purpose of emphasizing this need that the present work is published.

Undoubtedly many average loaf volumes considered significantly different should not be so regarded. The paramount importance of volume of the loaf in many past investigations in scoring flour for baking quality again appears to the authors to be over-emphasized. Sherwood and Bailey (1927), in comparing the standard baking test with the commercial loaf test, obtained a correlation of $+.824 \pm .047$ between the loaf volumes by the two methods. Later, on a more extensive series, Bailey, Fifield and Sherwood (1928) found the correlation to be only $+.53 \pm .07$. Undoubtedly these correlations have been lowered by molding and panning variability although that effect would be small since averages of 8 or 12 loaves were employed.

A correlation of but $+.53$ between volumes by two standard baking techniques indicates that approximately 85% of the variabilities of the entire series of loaf volumes by each method would be shown by that method for flours giving a constant volume by the other method. Thus loaf volumes resulting from a fixed baking technique are largely a reflection of that technique as applied to the flours and not of the flours themselves.

It may be well to consider what impression the baking replicates of this paper may have given if considered solely as pairs.

Bake series Nos. 1, 2, 3 and 8 are simple replicate series involving three workers and two standard flours, and are thus entirely suitable for this purpose. The average differences between successive members of these series are 14.5 cc., 11.6 cc., 12.1 cc. and 11.1 cc. respectively. The few pairs giving large differences of 30 cc. or more if increased to a trio by adding one of the immediately adjacent members of the series would show one "grossly in disagreement" with the other two. Of consecutive pairs of determinations in these four series of bakes, 55% agree within 10 cc. of each other, 72% within 15 cc. Yet the actual ranges of the entire series are 57 cc., 45 cc., 45 cc., and 50 cc., or approximately four times the average difference between consecutive replicates.

The ranges of small samples from normal populations have been rather fully discussed by Tippett (1925) and Pearson (1926), who have shown that samples of two may be expected to have an average difference of 1.128 times the standard deviation of the population from which they are drawn. Student (1927) has shown that the actual range is somewhat less than the theoretical range due to "secular variation" in replicate determinations. This is entirely in accordance with the present findings. It appears that the average difference between duplicate bakes may be expected to be approximately equal to the standard deviation of the distribution of a very large number of replicates. This difference is but one-fifth of the total range of 99 out of 100 replicates when smoothed by the normal curve. The fallacy of accepting the average difference between duplicates as a range beyond which deviations of volumes from different flours may be considered significant is obvious. Three times this magnitude for the minimum significant difference between volumes would represent only small chances in favor of differentiation of the flours.

Summary

Significantly different average loaf volumes of 627.5, 624.0 and 617.5 cc. were reported by three skilled workers baking 50 replicates each from the same flour in the same laboratory. Molding from doughs prepared by one worker alone indicated clearly that "molding personality" might well be the cause of these differences. It appears to be quite feasible that variation in molding technique may also be the cause of the variation in replicate volumes by any one worker, but such must remain an hypothesis until all other variables are controlled.

A squat type of pan produced loaves of significantly larger

volume and heavier weight than the specified tall pan of the standard baking procedure. The difference was much larger with a high grade flour than with one of low baking quality.

The absence of sugar from the dough of a standard flour resulted in loaves of approximately 1/6 lower volume but of good texture. The variability of the replicates was, however, doubled. The addition of 1 cc. of 0.1% $KBrO_3$ solution to the mix in the absence of sugar increased the loaf volume, reduced the loaf weight and reduced the variability of replicates.

The variability of the replicate loaf volumes due to other causes in routine experimental baking was such that fluctuation of the temperature of the dough within a range of 28° C. to 32° C. as it came from the mixing machine had no measurable effect upon the volume of the loaves in the present investigation.

The proofing cabinet was apparently uniform insofar as it affected loaf volume, since no place differentiation was demonstrable from the final loaf volumes in three pairs of analogous series of bakes.

Loaf volumes showed a systematically fluctuating average throughout the day's work for one baker, being more pronounced on certain days than on others. This was probably due either to fatigue or to the changing intensity of endeavor required by the work at different intervals of the day.

The average difference between duplicates in a large series of experimental bakes may be expected to be approximately equal to, or a little less than, the standard deviation of replicated loaf volumes. Thus, at least three times the average difference between duplicates should be taken as the minimum difference to be regarded as significant between volumes of single loaves baked from different flours.

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A STUDY OF COMMERCIALLY MILLED FLOURS DEALING WITH PROTEIN AND ITS RELATION TO PEPTIZATION AND BAKING STRENGTH

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The importance of peptization was first called to the attention of cereal chemists by Gortner, Hoffman and Sinclair (1927). They showed that the protein peptized from wheat flour varied inversely with the loaf volume. Harris (1930) reported results of a study involving experimentally milled flours, in which the relation of peptization of flour protein to loaf volume was investigated. Significant relationships were found between total protein and loaf volume and between protein peptizability and loaf volume.

The present study on commercially milled flours was done to ascertain whether these results, previously obtained by the author

on experimentally milled flours, would be confirmed by the use of commercial flours differing widely in type and origin.

A series of 31 flours, produced by mills in Western Canada, the Pacific Coast, and Central and Southeastern United States was studied. These flours were patents or straights, milled from different varieties of bread wheats, and grown under a variety of climatic and soil conditions. They were milled from the 1928 and 1929 wheat crops and included blends of both crops.

The baking procedure was as follows: the doughs were hand mixed in earthenware bowls and were run in pairs at intervals of 5 minutes. Fermentation, proofing time, and temperature were according to the Blish standard method (1928). The following formula was used:

Flour	100 grams
Yeast	3 grams
Sugar	2.5 grams
Salt	2 grams
Distilled water as required for proper consistency.	

The salt and sugar solution, as well as the water, were added from a burette. The yeast suspension was measured in a 10 cc. graduated cylinder. No attempt was made to use a constant absorption, as such a procedure would not have proved feasible for the range of moisture contents encountered with this series of flours. Owing to exposure to the air in sacks before being received at the laboratory, a number of the samples had lost much of their original moisture.

MacLeod (1929), Larmour and MacLeod (1929), and Larmour (1930), in a study of different series of Canadian wheat flours, found high relationships between total protein and baking strength when either bromate, malt, or Arkady and malt were included in the basic baking formula. When the basic formula was used without these improvers, these relationships were significantly lower. Harris (1930) also found a high relationship to exist between total protein, non-peptized protein and baking strength when these flour improvers were used. It was, therefore, thought advisable to include a baking formula containing these flour improvers, in addition to the basic procedure, in evaluating the strength of the flours of this series.

The improver used for the lower protein flours, including the majority of the first patents, was 1% diastatic malt and 0.001% $KBrO_3$. The malt was introduced as a solution, 12 cc. of which contained 3 g. of malt and 10 cc. of water. The $KBrO_3$ was added by

means of a pipette, 1 cc. of solution containing 0.001 g. of the chemical. Arkady and malt were used for the stronger flours, including the second patents and straights. The Arkady was added by means of a pipette, each 10 cc. representing 0.5 g. of improver. The malt solution was measured in a graduated cylinder. The treatment accorded the stronger flours would appear to correspond to that received in a commercial bakery, where Arkady and malt or similar improvers are commonly used.

In a previous paper on peptization, Harris (1930) reported a study of various methods. These methods included the procedure outlined by Gortner, Hoffman and Sinclair (loc. cit.), a method used by Larmour and MacLeod (1929), and a third method devised to obviate the use of a shaking machine and electric centrifuge. These three methods were found to give relative results and to be equally efficient for purposes of determining the peptizable protein. The third method, being the most convenient, was used for this study.

This procedure was as follows: 6 g. portions of the flour were weighed into 250 cc. Erlenmeyer flasks, 150 cc. of 0.5N $MgSO_4$ or KBr solution was added and the flour kept in suspension for one hour by frequent shaking. The suspended particles were allowed to settle, the supernatant liquid was decanted, aliquoted into two 50 cc. portions, and the nitrogen, extracted by the salt solution, determined by the Kjeldahl-Gunning procedure.

A description of the samples used in this study, with total protin, ash, and loaf volumes is given in Table 1. The first 15 flours shown in this table are first patents. Flour No. 101 had the highest protein content and produced the largest improver loaf volume of any member of this group. The flours Nos. 105 to 113 inclusive have much similar ash and protein contents. Their maximum difference in total protein is 0.7%, corresponding to a maximum difference in improver loaf volume of 70 cc. Samples Nos. 114 and 115 have the lowest protein contents of the first patents and they produced the lowest improver loaf volumes. The difference in average total protein of these two flours and No. 101 is 2.55%. The difference between the average basic loaf volumes for the same two flours and No. 101 is 28 cc., but when the improver values are considered, this difference increases to 103 cc. It would seem, therefore, that the improver method brings out more clearly the strength of the higher protein flour than the basic.

The group of second patents, Nos. 116 to 123, inclusive, show a tendency toward increasing improver loaf volume with increas-

ing protein content. This tendency is not so marked in the basic results, the first and highest protein member, No. 116, producing a basic loaf volume no larger than No. 123, the lowest protein flour of this group. Incidentally, No. 116, the second highest protein member of the 31 flours, produced the largest loaf, with malt and Arkady, of the entire series. Flours 119, 120 and 121 were very similar in ash, protein and improver loaf volume. These three flours showed a very strong positive response to malt and Arkady, yielding loaf volumes within 10 cc. of each other. The second patent group have higher total protein than the first patents.

The export flours, Nos. 124 to 127 inclusive, contained a large percentage of the lower quality mill streams, as is shown by the high ash content. Sample No. 127 was weaker than No. 124, and contained 3.4% less protein, corresponding to a difference of 150 cc. in improver loaf volume. The very high ash of No. 124, associated with dark loaf color, would keep this flour out of the class of baker's patents.

The last four flours of this series, Nos. 128 to 131 inclusive, were the lowest in total protein, and proved the weakest in baking strength. Flour No. 128 was the highest in protein of the four, and produced the largest loaf. The English wheat straight, No. 131, and the Pacific Club Patent, No. 130, were very similar in behaviour, both giving sticky doughs, the stickiness becoming more pronounced as fermentation progressed, necessitating the use of dusting flour. It will be noticed that these flours are both extremely low in protein, and gave low improver loaf volumes. When baked with malt and bromate included in the baking formula, these loaves were badly cracked and of poor crust color. Sample No. 129 was somewhat similar to these flours, but handled better, due probably to the higher protein content.

The greatest stimulation occasioned by the use of flour improvers was shown by the higher protein flours in the series of the 31 flours, such as Nos. 116, 117, 118, 119, 120 and 124. These flours had protein contents ranging from 13.5 to 15.1%, but yielded much the same loaf volumes with the basic method as lower protein flours. Flour No. 111, with a protein content of only 11.4%, produced a loaf by this method, of 540 cc. volume. The difference in protein between this flour and No. 116, is 3.2%, yet the difference in basic loaf volume is only 10 cc. With the use of a flour improver, however, the resultant loaf volumes of these two flours differ by 145 cc. These flours would be considered very similar in strength,

if judged by the loaves obtained by the use of the basic formula, when in reality one is very much stronger than the other.

No effort was made to compute a baking score for these flours, as the properties subject to investigation were concerned chiefly with loaf volume.

TABLE I

DESCRIPTION OF SAMPLE, ASH CONTENTS AND LOAF VOLUMES OF 31 COMMERCIALLY MILLED FLOURS

No.	Description	Total Protein	Ash	Loaf Volume	
				Basic	Improver
		%	%	cc.	cc.
	First Patents				
101	Canadian 1st Patent	13.2	0.35	525	610
102	Fancy Patent	12.3	.35	477	570
103	1st Patent Can. West	12.1	.40	505	565
104	1st Patent Can. West long extraction	12.0	.55	485	550
105	1st Patent Can. West	11.9	.42	525	572
106	1st Patent 1929 crop	11.8	.41	490	600
107	1st Patent Can. West	11.6	.43	505	575
108	1st Patent Can. West	11.5	.45	455	590
109	1st Patent Can. West	11.5	.43	440	550
110	1st Patent Can. West 1928	11.5	.42	500	530
111	1st Patent American Middle-West	11.4	.42	540	560
112	1st Patent Domestic crop 1928	11.3	.44	510	535
113	1st Patent Canadian West	11.2	.41	515	545
114	1st Patent Can. West, 1927	10.7	.39	500	490
115	1st Patent Can. West	10.6	0.42	495	525
	Second Patents				
116	2nd Patent Can. West, 1929	14.6	0.48	550	705
117	2nd Patent Can. West, 1929	13.7	.48	505	660
118	2nd Patent Can. West	13.6	.54	513	600
119	2nd Patent 1929 crop	13.6	.49	510	630
120	2nd Patent Can. West	13.5	.46	520	628
121	2nd Patent 1928 crop	13.4	.48	545	620
122	2nd Patent Can. West, 1929	12.7	.46	515	595
123	2nd Patent 1928 crop	12.2	0.48	550	625
	Export				
124	Export lower end of mill	15.1	0.68	510	680
125	Export 2nd Patent	12.2	.53	480	550
126	Straight—2nd Clear	12.7	.49	515	635
127	Export straight	11.7	0.54	490	530
	Weak				
128	Canadian small mill baker's flour	10.9	0.52	420	470
129	Soft Winter Patent	8.5	.43	430	445
130	Pacific Club Patent	7.8	.38	418	446
131	English Wheat Straight	7.0	0.45	440	450

Geddes and Goulden (1930) have shown that the relations between peptized protein, non-peptized protein and loaf volume depend largely upon the salts used for the peptization. When 0.5N $MgSO_4$ was used the non-peptized fraction was in general the more valuable portion but with 0.5N KI the peptized pro-

tein was the more valuable in relation to baking quality. They also stated that correlations computed between percentage of total protein peptized and loaf volume do not accurately measure the relative value of the peptized and non-peptized fractions for baking purposes, since their magnitude reflects, in part, the relation between total protein and loaf volume. Similar conclusions were also reached by Harris (loc. cit.). It therefore seemed advisable in view of these results to calculate the peptized and non-peptized fractions in the present series.

Values of these fractions, together with loaf volume, total protein, and peptized protein as per cent of the total, are shown in Table II.

A slight increase in per cent total protein peptized is evident for both salt solutions in the first patent group, Nos. 101 to 116 inclusive, in proceeding from the higher protein to the lower protein flours. No great differences in per cent total protein extracted are noted in the second patent or export flours, but the weaker flours at the end of the series give significantly higher values.

Peptized protein does not appear to vary greatly in any of the classes. Non-peptized protein shows a steady decrease for each salt solution in all flour classes with decreasing protein content, reaching a minimum in the soft wheat flours.

The data presented in this table were subjected to a statistical analysis according to the method developed by Geddes and Goulden (loc. cit.). The various correlations obtained are shown in Table III. The partial correlations were employed to measure the effect of one protein fraction upon loaf volume without the interference of the other fraction. The multiple correlations were employed to show the combined effect of the peptized and non-peptized fractions upon loaf volume.

The correlations obtained for the two baking methods show more significant relationships when an improver was used. This finding is in accord with the work reported previously by the author when a series of experimental flours was investigated. Total and non-peptized protein appeared to be of approximately equal importance in predicting loaf volume, whereas per cent total protein peptized is much less significant. The peptized fraction apparently does not have a relationship of any great practical importance with the loaf volumes of this series of flours. The high relationships obtained by correlating peptized protein and improver loaf volume in the KBr group are evidently due to the influence of the non-peptized fraction, as the relationship between

these variables is scarcely significant when the interference of non-peptized protein is removed by partial correlation. This conclusion is strengthened by the correlation shown between the protein fractions for KBr.

Non-peptized protein for KBr shows a slightly less signficant relationship with loaf volume when the effect of peptized protein is removed by partial correlation.

Figure 1 shows a fairly regular increase of loaf volume with increasing protein and corresponds very closely to a similar figure for 44 experimentally milled samples discussed in a previous paper. In Figure 2, there is more scattering than in Figure 1, showing less probability of predicting loaf volume from protein content. Figures 3 and 4 show the relationships between non-peptized protein and loaf volume and are a reflection, to a large extent, of Figures 1 and 2. They show no greater practical value in forecasting baking strength from a knowledge of non-peptized protein than from a knowledge of total protein.

I. Millstream Flours

In order to determine whether the conclusions derived from this previous study would be confirmed by the use of a series of flours milled from the same wheat blend, twenty millstream flours were investigated in the same manner as the group of 31 commercial flours described above.

These flours were peptized by the same method used for the commercial flours and were analyzed and baked. Two methods of baking were employed, the basic and one containing 3% malt and 0.5% Arkady in addition to the basic ingredients.

A baking score was computed for these flours, in the following manner:

Loaf volume	x 0.1		
Symmetry	x 1.0	Maximum value	10
Grain of loaf	x 1.0	Maximum value	10
Color	x 1.0	Maximum value	20
Texture	x 1.0	Maximum value	10

The sum of these individual scores was considered the baking value of the flour.

A description of the millstream flours used in this study is shown in Table IV, including ash, protein and moisture contents of these flours, loaf volumes and scores. A range of 8.8% in protein is shown among these samples, and a difference of 490 cc. between the largest and smallest loaves. The baking score ranges from 37.2 to 123.

TABLE II

COMPARATIVE BAKING AND PEPTIZATION DATA FOR 31 COMMERCIALLY MILLED FLOURS

No.	Total Protein as Per Cent of Flour	Loaf Volume		KBr			$MgSO_4$		
		Basic	Improver	Total Protein Peptized	Peptized Protein	Non-peptized Protein	Total Protein Peptized	Peptized Protein	Non-peptized Protein
	%	cc.	cc.	%	%	%	%	%	%
				First Patents					
101	13.2	525	610	27.2	3.59	9.61	15.0	1.98	11.22
102	12.3	477	570	27.5	3.38	8.92	15.1	1.85	10.45
103	12.1	505	565	27.2	3.29	8.81	16.1	1.95	10.15
104	12.0	485	550	30.6	3.67	8.33	17.5	2.10	9.90
105	11.9	525	572	28.1	3.34	8.56	16.5	1.96	9.94
106	11.8	490	600	29.2	3.44	8.36	19.3	2.28	9.52
107	11.6	505	575	30.2	3.50	8.10	16.3	1.89	9.71
108	11.5	455	590	29.3	3.37	8.13	19.8	2.28	9.22
109	11.5	440	550	30.1	3.46	8.04	17.9	2.06	9.44
110	11.5	500	530	30.9	3.55	7.95	17.9	2.06	9.44
111	11.4	540	560	27.2	3.10	8.30	17.6	2.00	9.40
112	11.3	510	535	29.9	3.38	7.92	19.6	2.21	9.09
113	11.2	515	545	30.0	3.36	7.84	17.0	1.90	9.30
114	10.7	500	490	27.5	2.94	7.76	17.3	1.85	8.85
115	10.6	495	525	31.5	3.34	7.26	18.4	1.95	8.65
				Second Patents					
116	14.6	550	705	28.2	4.12	10.48	16.5	2.40	12.20
117	13.7	505	660	26.9	3.68	10.02	16.6	2.27	11.43
118	13.6	513	600	29.2	4.97	8.63	16.5	2.24	11.36
119	13.6	510	630	27.9	3.79	9.81	17.9	2.43	11.17
120	13.5	520	628	27.1	3.66	9.84	16.9	2.28	11.22
121	13.4	545	620	27.3	3.66	9.74	16.8	2.25	11.15
122	12.7	515	595	28.2	3.58	9.12	17.7	2.25	10.45
123	12.2	550	625	28.9	3.53	8.67	19.0	2.32	9.88

Note—peptized and non-peptized protein calculated as per cent of flour.

TABLE II—(Continued)

COMPARATIVE BAKING AND PEPTIZATION DATA FOR 31 COMMERCIALLY MILLED FLOURS

No.	Total Protein as Per Cent of Flour	Loaf Volume		KBr			$MgSO_4$		
		Basic	Improver	Total Protein Peptized	Peptized Protein	Non-peptized Protein	Total Protein Peptized	Peptized Protein	Non-peptized Protein
	%	cc.	cc.	%	%	%	%	%	%
				Export					
124	15.1	510	680	28.5	4.29	10.81	17.1	2.58	12.52
125	12.2	480	550	29.2	3.56	8.64	18.6	2.27	9.93
126	12.7	515	635	29.1	3.70	9.00	17.0	2.16	10.54
127	11.7	490	530	29.9	3.50	8.20	19.5	2.28	9.42
				Weak					
128	10.9	420	470	30.5	3.32	7.58	19.6	2.14	8.76
129	8.5	430	445	32.5	2.76	5.74	20.6	1.75	6.75
130	7.8	418	446	32.4	2.53	5.27	23.4	1.83	5.97
131	7.0	440	450	36.8	2.58	4.42	26.4	1.85	5.15

Note—peptized and non-peptized protein calculated as per cent of flour.

It will be noticed that the first three middling flours are the lowest in ash and protein, and produce loaves yielding the highest color and grain scores. These flours originate from the central portion of the endosperm, which contains the smallest quantity of nitrogenous and inorganic material, and is the most suitable for first patent, highly refined flour. The fourth and fifth middlings are appreciably higher in ash and produced bread of poorer grain. It is generally considered that these flours come from a portion of the wheat berry external to the origin of the three streams first

TABLE III

STATISTICAL ANALYSIS OF 31 COMMERCIALLY MILLED FLOURS

		$MgSO_4$		KBr	
		Basic	Improver	Basic	Improver
r_{ae}	=	$+ .6918 \pm .0631$	$+ .9125 \pm .0203$	$+ .6918 \pm .0631$	$+ .9125 \pm .0203$
r_{ad}	=	$- .7872 \pm .0461$		$- .7925 \pm .0450$	
r_{be}	=	$+ .1107 \pm .1196$	$+ .2023 \pm .1161$	$+ .5269 \pm .0875$	$+ .7538 \pm .0523$
r_{bc}	=	$+ .1865 \pm .1169$		$+ .7546 \pm .0521$	
r_{ce}	=	$+ .7052 \pm .0609$	$+ .9027 \pm .0224$	$+ .7033 \pm .0612$	$+ .9085 \pm .0211$
r_{de}	=	$- .6327 \pm .0726$	$- .6319 \pm .0727$	$- .6633 \pm .0678$	$- .6830 \pm .0646$
${}_br_{ce}$	=	$+ .7012 \pm .0616$	$+ .8989 \pm .0252$	$+ .5481 \pm .0848$	$+ .7878 \pm .0459$
${}_cr_{be}$	=	$- .0298 \pm .1210$	$+ .0804 \pm .1203$	$- .0081 \pm .1211$	$+ .2491 \pm .1136$
$R_{(bc)e}$	=	$+ .7054 \pm .0608$	$+ .9032 \pm .0223$	$+ .7029 \pm .0613$	$+ .9146 \pm .0198$

NOTE:—

a = total protein
b = peptized protein
c = non-peptized protein
d = % total protein peptized
e = loaf volume

described. The loaf volume and total protein are much similar to the first three middlings flours.

In the break flours, a large increase in protein is evident, corresponding to a high loaf volume. The change in loaf volume is especially striking when the improver was used. The ash in this group is not much higher than in the fourth and fifth middling flours, but the color score shows a progressive decrease. These break flours originate much nearer the exterior of the berry than the middlings flour, although portions of the endosperm proper, as well as germ particles, find their way into these streams.

The sizings flours are much alike in protein content and volume of loaf. The ash content, however, is much higher in the second sizings, associated with a poor loaf color. The break cuts is a stream much similar to the break flours in origin and characteristics. The remainder of the flours become progressively poorer in quality as the end of the system is approached. The fourth and

fifth break flours contain very high percentages of protein, but appear unable to yield loaf volumes as high as the first three break flours under the action of an improver. The ash content is high, and the color score low, indicating presence of material from the vicinity of the bran. The inclusion of such material is also shown in the high moisture content of these flours as the outer portion of the berry would contain the greater proportion of the tempering

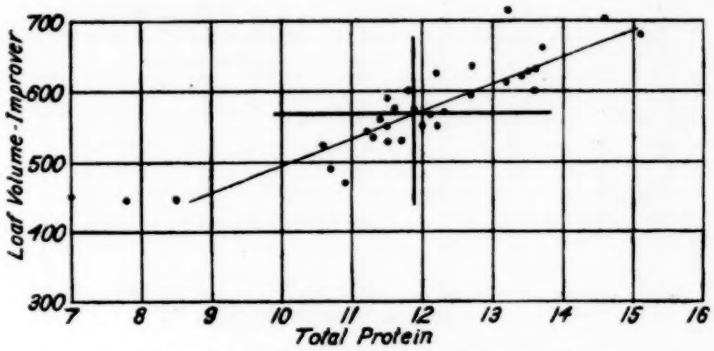

Fig. 1. Total Protein (Per Cent) and Loaf Volume (Improver) for 31 Commercial Flours
$r_{ae} = +.9125 \pm .0203$

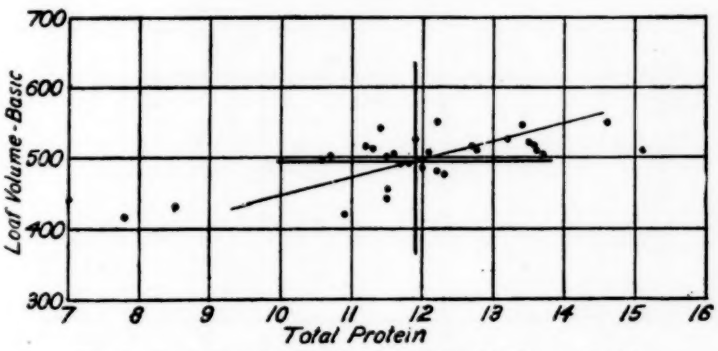

Fig. 2. Total Protein (Per Cent) and Loaf Volume (Basic) for 31 Commercial Flours
$r_{ae} = +.6918 \pm .0631$

water. The reel flours are very high in ash, and yielded loaves of very poor color, this being especially true of reel No. 4. A progressive loss of moisture is evident as the stocks pass through the mill, due to evaporation of water from the flour streams. The breaks are the highest in moisture content, while streams such as tailings, 8th middlings and the reels, are the lowest.

The peptization data obtained for these flours, calculated as peptized protein, non-peptized protein and per cent of total protein

peptized, are shown in Table V. 0.5N KI was included to ascertain the effect of its high peptizing power upon this series of flours. The loaf volumes and total flour proteins are also given in this table to facilitate comparison.

Great variability is evident in the percentage of total protein

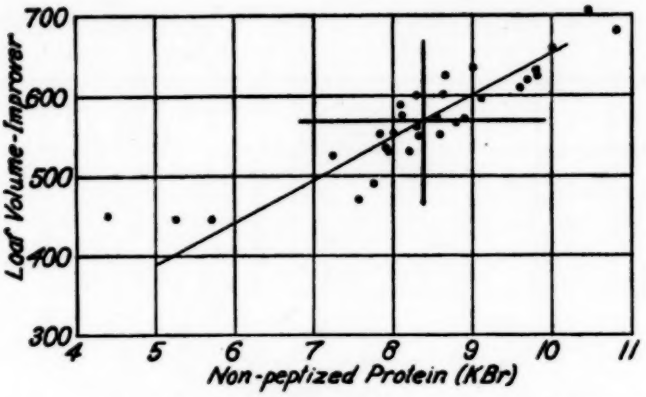

Fig. 3. Non-peptized Protein (KBr) and Loaf Volume (Improver) for 31 Commercial Flours
$r_{ce} = .9085 \pm .0211$

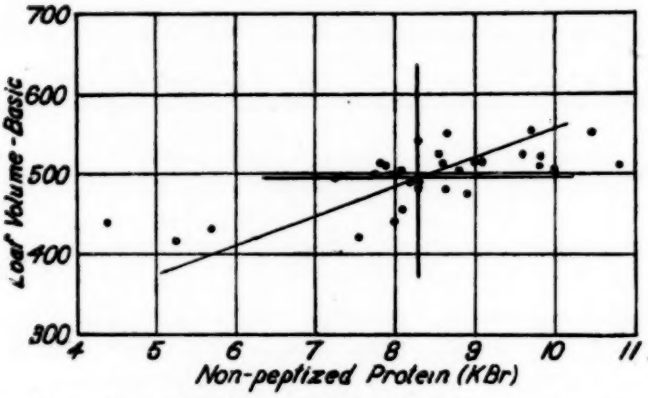

Fig. 4. Non-peptized Protein (KBr) and Loaf Volume (Basic) for 31 Commercial Flours
$r_{ce} = .7033 \pm .0612$

extracted from the different flours by any one salt solution. The $MgSO_4$ values range from 12.5 to 40.9; the KBr values from 22.2 to 45.4; the KI values from 44.0 to 63.6. All these salt solutions gave very high relative values for reel No. 4. The break flours yielded consistently lower results, associated with large loaf volumes and high total protein. The first three breaks produced exceptionally large volumes when treated with improver, but the fourth and fifth

break flours gave a much inferior response to the action of malt and Arkady in spite of the fact that they contained considerably

TABLE IV

COMPARATIVE BAKING AND ANALYTICAL DATA FOR 20 MILLSTREAM FLOURS
(Protein, ash and loaf volume calculated to 13.5% moisture basis)

Flour	Moisture	Protein	Ash		Loaf Volume	Symmetry	Color	Grain	Texture	Score
	%	%	%							
				B[1]	480	8	15	9	9.5	79.5
1st Midds.	13.7	12.3	.42	I[2]	580	8	17	9	8	100
				B	465	8	15	9.5	9.5	88.5
2nd Midds.	13.7	12.2	.39	I	565	10	17	9	9	101.5
				B	462	9	16	9	9.5	89.7
3rd Midds.	13.0	11.6	.34	I	530	10	17.5	9	8.5	98
				B	490	8	11	8	9	85
4th Midds.	13.5	12.9	.54	I	600	9	12	8	7	106
				B	465	8	12	8	9	83.5
5th Midds.	13.4	12.3	.47	I	580	9	10	8.5	7.5	92
				B	520	9	10	7	8	86
1st Break	14.3	15.1	.64	I	760	8	13	8	8	113
				B	555	10	10.5	8	8	92
2nd Break	14.3	15.6	.45	I	835	11	12	8	8	123
				B	560	10	10.5	8	8	92.5
3rd Break	13.7	17.6	.49	I	840	12	11	8	8	123
				B	485	6.5	13	6	7	81
1st Sizings	14.1	12.2	.47	I	560	7	9	6	7	85
				B	485	6.5	10	6	6	77
2nd Sizings	13.7	12.8	.60	I	550	7	7	6.5	6	81.5
				B	542	10	10	5	5	84.2
Break Cuts	14.8	15.6	.47	I	780	12	12	7	8	117
				B	480	6	10	7	6	77
6th Midds.	12.3	12.7	.61	I	555	8	11	7	6	87.5
				B	450	5	8	4	4	66
7th Midds.	12.8	13.8	.76	I	572	8	9	4	4	82.2
				B	400	3	5	2	3	53
8th Midds.	12.3	14.3	1.18	I	435	4	5	3	3	58.5
				B	545	5	8	5	6	78.5
4th Break	14.8	19.2	.57	I	695	10	10	4	5	98.5
				B	565	4.5	6	4	5	86
5th Break	14.3	20.4	.83	I	660	9.5	7	4	4	90.5
				B	500	7	8	7	7	79
1st Tailing	12.7	15.8	.73	I	645	9	9	6	5	93.5
				B	465	6	7	5	6	70.5
2nd Tailing	12.6	14.6	.82	I	580	8	7.5	6	5	84.2
				B	530	7	6	7	5	78
Reel No. 2	13.4	18.3	1.05	I	610	9	7	5	4	86
				B	332	0	1	2	1	37.2
Reel No. 4	11.4	16.3	1.92	I	350	1	2	3	2	43

[1] Basic
[2] Improver

more protein than the first three break flours. It would appear that the improver revealed a weakness in these two flours, not shown by the basic baking method.

The proteolytic activity of various flour streams in the Minnesota State Mill was determined by Bailey and Cairns (1928). They found the fourth and fifth breaks remarkably high in this respect. Geddes (1929) found that when a flour was heat treated to 150° F., no injury was apparent by the basic method, but, rather, an improvement was noted. Treatment by bromate, however, revealed damage in all cases. Larmour and MacLeod (loc. cit.) showed that weakness in many wheats escaped detection until an improver was included in the baking formula. It might be that protease activity had weakened the fourth and fifth break flours of the present study sufficiently for this to be revealed by treatment with a strong improver, such as 3% malt and 0.5% Arkady.

Non-peptized protein appears to be a reflection of total protein, except in the lower grade flour streams, where the non-peptized value is lower than would be expected, if judged by the protein content of the flour. Peptized protein is higher for these poor samples as compared with the stronger flours.

For purposes of statistical study, the data obtained from the 20 millstream flours was divided into two groups. The first group comprises 12 flour streams, corresponding in their action to the 31 commercial flours, and also to the 44 experimental flours discussed in a previous paper. This group includes the first six middlings, the first three breaks, first and second sizings and break cut flours. A statistical analysis of these 12 flours is given in Table VI. The other group includes the data from the entire 20 flours, as it was desired to determine the effect of the lower quality flour streams upon the relations of total protein and protein peptization to loaf volume. A statistical treatment of this data is given in Table VII.

The use of the improver did not yield correlations significantly higher than the basic method for the 12 flour streams as shown in Table VI. Therefore, no further information was gained by including malt and Arkady in the basic formula. This result is contrary to the conclusions reached for the experimental and commercial flours studied. Very significant correlations were shown between total protein and loaf volume for these 12 flours. There were also high correlations between non-peptized protein and loaf volume, these correlations becoming smaller in the KI group, however, when the influence of the peptized protein was removed by partial correlation. These relationships between total protein and loaf volume, and between non-peptized protein and loaf volume, would appear to be of sufficient importance to justify forecasting baking

TABLE V

TOTAL PROTEIN, LOAF VOLUMES AND PEPTIZED AND NON-PEPTIZED PROTEIN FOR TWENTY MILLSTREAM FLOURS

Flour	Loaf Volumes			$MgSO_4$			KBr			KI		
	Total Protein as % of Flour	Basic	Improver	Total Protein Peptized	Peptized Protein as % of Flour	Non-Peptized Protein as % of Flour	Total Protein Peptized	Peptized Protein as % of Flour	Non-Peptized Protein as % of Flour	Total Protein Peptized	Peptized Protein as % of Flour	Non-Peptized Protein as % of Flour
	%	cc.	cc.	%	%	%	%	%	%	%	%	%
1st Middlings	12.3	480	580	16.4	2.02	10.28	28.5	3.51	8.79	49.4	6.08	6.22
2nd Middlings	12.2	465	565	16.4	2.00	10.20	28.4	3.46	8.74	51.7	6.31	5.89
3rd Middlings	11.6	462	530	16.8	1.95	9.65	28.0	3.25	8.35	50.5	5.86	5.74
4th Middlings	12.9	490	600	17.4	2.24	10.66	28.8	3.72	9.18	52.6	6.79	6.11
5th Middlings	12.3	465	580	17.9	2.20	10.10	32.3	3.97	8.33	50.7	6.23	6.07
1st Break	15.1	520	760	15.2	2.30	12.80	24.4	3.68	11.42	48.6	7.34	7.76
2nd Break	15.6	555	835	12.5	1.95	13.65	25.3	3.95	11.65	47.7	7.44	8.16
3rd Break	17.6	560	840	13.2	2.32	15.28	23.6	4.15	13.45	46.9	8.25	9.35
1st Sizing	12.2	485	560	16.0	1.95	10.25	27.4	3.34	8.86	51.0	6.22	5.98
2nd Sizing	12.8	485	550	18.0	2.30	10.50	29.9	3.83	8.97	52.2	6.68	6.12
Break Cuts	15.6	542	780	15.1	2.36	13.24	26.4	4.12	11.48	50.0	7.80	7.80
6th Middlings	12.7	480	555	19.8	2.51	10.19	31.0	3.93	8.77	51.3	6.52	6.18
7th Middlings	13.8	450	572	19.8	2.73	11.07	32.8	4.53	9.27	53.6	7.40	6.40
8th Middlings	14.3	400	435	28.8	4.12	10.18	38.0	5.43	8.87	59.4	8.49	5.81
4th Break	19.2	545	695	14.3	2.75	16.45	22.2	4.26	14.94	47.1	9.04	10.16
5th Break	20.4	565	660	15.9	3.74	16.66	23.9	4.88	15.52	47.4	9.67	10.73
1st Tailing	15.8	500	645	16.8	2.65	13.15	27.9	4.41	11.39	52.1	8.23	7.57
2nd Tailing	14.6	465	580	19.4	2.83	11.77	29.3	4.28	10.32	48.8	7.12	7.48
Reel No. 2	18.3	530	610	16.6	3.04	15.26	23.3	4.26	14.04	44.0	8.05	10.25
Reel No. 4	16.3	332	350	40.9	6.67	9.63	45.4	7.40	8.90	63.6	7.58	8.72

strength in the flours of this series from a knowledge of total or non-peptized protein. The correlations between per cent total protein peptized and loaf volume, while quite significant, show less relationship than the corresponding values for total and non-peptized protein. The correlation $_{c}r_{be}$ does not reveal as high relations between peptized protein and loaf volume as exist between non-peptized protein and loaf volume. This relationship, however, tends to increase algebraically in going from $MgSO_4$ to KI: for example, the correlation for $MgSO_4$ and improver loaf volume is $-.5419$, while for KI it is $+.1509$. These conclusions, with the exception of that dealing with the baking method, correspond to the conclusions reached from a statistical study of the data obtained from the experimental and commercial flours previously reported by the author.

The data presented in Table VII, shows somewhat different relationships between the variables discussed. The inclusion of malt and Arkady in the baking formula appeared to reduce the significance of the correlations concerned with baking strength. Total protein was much less significantly related to loaf volume than in the former series of flours and would not seem to be of any practical value in predicting baking strength. The inclusion of data from high protein, low quality streams has affected this relationship in this series of flours. Non-peptized protein was more highly correlated than total protein in respect to baking strength but these values were lower than the corresponding values mentioned in Table VI. These relationships are much lower for the KI group but are quite high for the KBr series when the influence of peptized protein is removed by partial correlation. The highest correlation shown in Table VII is between per cent total protein peptized by KI and loaf volume. The non-peptized as well as the multiple values are highest in the $MgSO_4$ series. The correlation $_{c}r_{be}$ given in this table shows a general tendency to increase algebraically from $MgSO_4$ to KI. A similar trend was noted in the correlations shown in Table VI. These tendencies appear to support the "optimum coagulation theory" of Kent-Jones.

In Table VIII the statistical constants computed from the data obtained by the author for the 44 experimental flours are repeated for purposes of comparison with the values obtained in the present study.

An analysis of variance was applied to determine the relative significance of the correlation coefficients r_{ae} and $R_{(bc)e}$ reported in this study. The two coefficients cannot be compared di-

TABLE VI

STATISTICAL TREATMENT OF THE DATA OBTAINED FROM 12 MILLSTREAM FLOURS OF PATENT QUALITY

		$MgSO_4$		KBr		KI	
		Basic	Improver	Basic	Improver	Basic	Improver
r_{ae}	=	$+.9653 \pm .0133$	$+.9631 \pm .0141$	$+.9653 \pm .0133$	$+.9631 \pm .0141$	$+.9653 \pm .0133$	$+.9631 \pm .0141$
r_{ad}	=	$-.7583 \pm .0827$		$-.6772 \pm .1054$		$-.7766 \pm .0773$	
r_{be}	=	$+.2326 \pm .1842$	$+.1848 \pm .1880$	$+.6536 \pm .1115$	$\pm .6343 \pm .1164$	$+.9441 \pm .0212$	$+.9226 \pm .0290$
r_{bc}	=	$+.3078 \pm .1763$		$\pm .6163 \pm .1207$		$+.9409 \pm .0223$	
r_{ce}	=	$+.9719 \pm .0108$	$+.9747 \pm .0097$	$+.9632 \pm .0141$	$+.9642 \pm .0137$	$+.9564 \pm .0166$	$+.9666 \pm .0128$
r_{de}	=	$-.8118 \pm .0664$	$-.8453 \pm .0556$	$-.7987 \pm .0705$	$-.8063 \pm .0672$	$-.7599 \pm .0823$	$-.7705 \pm .0791$
$^{b}r_{ce}$	=	$+.9729 \pm .0104$	$+.9816 \pm .0071$	$+.9403 \pm .0225$	$+.9416 \pm .0221$	$+.6097 \pm .1223$	$+.7536 \pm .0841$
$^{c}r_{be}$	=	$-.2968 \pm .1775$	$-.5419 \pm .1375$	$+.2834 \pm .1791$	$+.1920 \pm .1875$	$+.4469 \pm .1558$	$+.1509 \pm .1903$
$R_{(bc)e}$	=	$+.9743 \pm .0099$	$+.9821 \pm .0069$	$+.9661 \pm .0130$	$+.9655 \pm .0132$	$+.9658 \pm .0131$	$+.9672 \pm .0126$

NOTE:— a = Total Protein; b = Peptized Protein; c = Non-Peptized Protein; d = % Total Protein Peptized; e = Loaf Volume

TABLE VII

STATISTICAL TREATMENT OF THE DATA OBTAINED FROM 20 MILLSTREAM FLOURS

		$MgSO_4$		KBr		KI	
		Basic	Improver	Basic	Improver	Basic	Improver
r_{ae}	=	$+.4538 \pm .1197$	$+.3928 \pm .1275$	$+.4538 \pm .1197$	$+.3928 \pm .1275$	$+.4538 \pm .1197$	$+.3928 \pm .1275$
r_{ad}	=	$-.0435 \pm .1505$		$-.2426 \pm .1419$		$-.3077 \pm .1365$	
r_{be}	=	$-.6006 \pm .0964$	$-.5516 \pm .1049$	$-.5557 \pm .1042$	$-.4484 \pm .1205$	$+.3427 \pm .1331$	$\pm .2958 \pm .1376$
r_{bc}	=	$-.0240 \pm .1507$		$+.0901 \pm .1496$		$+.7947 \pm .0557$	
r_{ce}	=	$+.7757 \pm .0601$	$+.6858 \pm .0799$	$+.7220 \pm .0722$	$+.6117 \pm .0932$	$+.4869 \pm .1150$	$+.4214 \pm .1240$
r_{de}	=	$-.8848 \pm .0327$	$-.7845 \pm .0580$	$-.9439 \pm .0164$	$-.7995 \pm .0544$	$-.8838 \pm .0330$	$-.7272 \pm .0710$
$^{b}r_{ce}$	=	$+.9524 \pm .0140$	$+.8065 \pm .0527$	$+.9324 \pm .0197$	$+.7326 \pm .0699$	$+.3763 \pm .1294$	$+.3363 \pm .1337$
$^{c}r_{be}$	=	$-.9225 \pm .0225$	$-.7354 \pm .0692$	$-.8159 \pm .0504$	$-.6390 \pm .0892$	$-.0833 \pm .1498$	$-.0710 \pm .1500$
$R_{(bc)e}$	=	$+.9697 \pm .0090$	$+.8699 \pm .0367$	$+.9537 \pm .0136$	$+.7934 \pm .0559$	$+.4939 \pm .1140$	$+.4267 \pm .1233$

NOTE:— a = % Total Protein Peptized; b = Loaf Volume; c = Peptized Protein; d = Non-Peptized Protein; e = Total Protein

rectly, as they were calculated from the same data, and might themselves be correlated. This method was used by Geddes and Goulden (loc. cit.) for a similar purpose, and it is based upon Fisher's method of applying the analysis of variance to determin-

TABLE VIII

STATISTICAL TREATMENT OF THE DATA OBTAINED FROM 44 EXPERIMENTALLY MILLED FLOURS

		$MgSO_4$		KBr	
		Basic	Improver	Basic	Improver
r_{ae}	=	$+ .6937 \pm .0527$	$+ .9011 \pm .0191$	$+ .6937 \pm .0527$	$+ .9011 \pm .0191$
r_{ad}	=	$- .6209 \pm .0625$		$- .7165 \pm .0495$	
r_{be}	=	$+ .3908 \pm .0862$	$+ .3866 \pm .0865$	$+ .3667 \pm .0880$	$+ .2585 \pm .0949$
r_{bc}	=	$+ .3014 \pm .0925$		$+ .2390 \pm .0959$	
r_{ce}	=	$+ .6735 \pm .0556$	$+ .8921 \pm .0208$	$+ .6640 \pm .0569$	$+ .9128 \pm .0171$
r_{de}	=	$- .4789 \pm .0784$	$- .6148 \pm .0632$	$- .4579 \pm .0804$	$- .7326 \pm .0471$
${}_b r_{ce}$	=	$+ .6331 \pm .0609$	$+ .8820 \pm .0226$	$+ .6379 \pm .0603$	$+ .9073 \pm .0181$
${}_c r_{be}$	=	$+ .2664 \pm .0945$	$+ .2732 \pm .0941$	$+ .2863 \pm .0932$	$+ .1015 \pm .1006$
$R_{(bc)e}$	=	$+ .7017 \pm .0725$	$+ .9006 \pm .0193$	$+ .6875 \pm .0536$	$+ .9132 \pm .0169$

NOTE:—

a = Total Protein
b = Peptized Protein
c = Non-Peptized Protein
d = % Total Protein Peptized
e = Loaf Volume

TABLE IX

COMPARISON BY ANALYSIS OF VARIANCE OF TOTAL AND MULTIPLE CORRELATION COEFFICIENTS

Correlation Coefficient	$MgSO_4$		KBr		KI	
	Basic	Improver	Basic	Improver	Basic	Improver
Commercial Flours						
r_{ae}	.6918	.9125	.6918	.9125		
$R_{(bc)e}$	.7054	.9032	.7029	.9146		
z	.0298+	.4701+	.0748−	.2199−		
Value of z at 5% point, .7171						
Twelve Millstream Flours						
r_{ae}	.9653	.9631	.9653	.9631	.9653	.9631
$R_{(bc)e}$	.9743	.9821	.9661	.9655	.9658	.9672
	.5698+	1.1082+	.7657+	.2444−	1.0007−	.0464+
Value of z at 5% point, .8163						
Twenty Millstream Flours						
r_{ae}	.4538	.3928	.4538	.3928	.4538	.3928
$R_{(bc)e}$	.9697	.8699	.9537	.7934	.4939	.4267
z	2.6731+	1.8703+	2.4443+	1.5409+	.0923−	.2741
Value of z at 5% point, .7446						

ing the significance of regression coefficients. Results of this test are summarized in Table IX. A significant increase of the multiple over the total correlation is shown by a z value of .7171 for 31

samples, and in the other series according to the degrees of freedom. A negative sign placed after the z value indicates a loss of information, and the table of z must be entered in the other direction. There are no indications of a significant loss of information in this table when the multiple correlations are considered instead of the simple values.

Inspection of the table reveals the fact that apparently the usefulness of a knowledge of the protein fractions is greatest for the $MgSO_4$ and KBr extractions in the 20 millstream series. This is probably associated with less relationship between total protein and loaf volume. No indication of a significant gain of information is shown in the other two sets of constants, except for $MgSO_4$ and improver loaf volume for the 12 millstreams.

The correlations found for the millstreams in the present study are much higher than those reported by Pascoe, Sherwood and Gortner (1930) between loaf volume, total protein and per cent total protein peptized by various salts for a series of 17 millstream flours. They found the correlation between protein content and loaf volume to be $r = -.018 \pm .164$. The correlation found between the peptizable protein fraction and loaf volume for KF was $r = -.317 \pm .147$; for KCl, $r = -.235 \pm .072$; for KBr, $r = -.186 \pm .158$ and for $MgSO_4$, $r = -.241 \pm .154$.

Summary

A series of 31 commercially milled flours of different baking strengths was baked and analyzed. The proteins of these flours were peptized by 0.5N solutions of $MgSO_4$ and KBr. Two methods of baking were used: the basic method employing flour, sugar, salt, yeast and water, and a method which included 1% malt and 0.001% $KBrO_3$ for the weaker flours, and 3% malt and 0.5% Arkady for the stronger flours, in addition to the basic formula.

The data obtained was subjected to a statistical analysis. The improver loaf volume showed significantly higher relationships than the basic loaf volume when correlated with the other variables. This appears to demonstrate the advisability of using a flour improver in evaluating the baking strength of commercial flours.

Total and non-peptized protein gave very high relationships when correlated with loaf volume and would appear to be equally useful as a means of forecasting baking strength for this series.

Per cent total protein peptized was less significantly related to loaf volume than total or non-peptized protein and would be of less value in predicting baking strength.

Peptized protein did not appear to be of any great practical importance in its relation to baking strength.

These conclusions are similar to those obtained in a previous study on experimentally milled flours by the author.

Further, a series of 20 millstream flours was baked, by two methods, the basic and one including 3% malt and 0.5% Arkady. These flours were also analyzed, and peptized by 0.5N $MgSO_4$, KBr and KI. The resultant data was considered in two groups, the streams which appeared to be of good baking quality, comprising 12 flours, were grouped in one series while the entire 20 streams, including low grade flour streams, were placed in another. The data in both these groups were statistically treated in the same way as the data obtained from the 31 commercially milled flours.

The correlations involving the basic and improver baking methods were equally significant for the group of 12 millstream flours, while for the 20 millstreams the basic procedure showed higher relationships. This does not appear to justify the use of an improver in evaluating the strength of millstream flours.

Total protein and loaf volume were highly correlated for the 12 flours but these variables showed a relation of no great practical importance in the group of 20, due to the inclusion of high protein, low quality flours. It would appear from this that total protein is not a reliable index of baking strength when low grade millstreams are concerned.

Non-peptized protein and loaf volume showed a high relationship for the group of 12 flours, this relationship decreasing when the poorer flour streams were included in the study. This last correlation was higher than the corresponding value for total protein and loaf volume. From this it would seem that non-peptized protein is more significant in its relation to baking strength than total protein, in millstream flours including the low grade streams

Per cent total protein peptized appears to be equal to or greater than total protein or non-peptized protein for purposes of predicting baking strength of millstream flours which include the lower grade streams.

The "optimum coagulation" theory appears to be supported by the peptization data from the millstream flours.

A consideration of the amount of peptized and non-peptized protein in addition to total protein, yielded little additional information in respect to loaf volume, except for $MgSO_4$ and KBr for the entire set of millstream flours. This is probably due to a lower correlation between total protein and loaf volume in this series.

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WHEAT AND FLOUR STUDIES XVIII. A STUDY OF THE NATURE OF THE ACID RESPONSIBLE FOR THE INCREASE IN ACIDITY WHICH OCCURS IN FLOURS DURING STORAGE[1]

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It has been known for a long time that flours increase in their acidity during storage. This increased acidity has been attributed to the operation of several different phenomena: gradual hydrolysis of the fat to yield fatty acids; hydrolysis of protein to yield amino acids, or intermediate products of protein decomposition; and enzymic splitting of phytin to yield phosphoric acid compounds. Jessen-Hansen (1911) was the first to express the increasing acidity of stored flours in terms of the H-ion concentration of their extracts and to show that flours increased in their H-ion concentration during storage. Sharp (1924) and Bailey and Johnson (1924) presented further data in support of Jessen-Hansen's observation. Bailey and Johnson also noted that a freshly milled flour, extracted with ether, showed practically no change in H-ion concentration during storage for 3 years. This has led them to ask whether the ether removed from the flour the material which contributed to the increasing acidity of normal flour or whether the ether inactivated the organisms or enzymes instrumental in modifying the H-ion concentration. While the first alternative seemed the more probable to them, the present work has been done with the idea of establishing it more certainly and of determining the nature of the acids responsible for the change in H-ion concentration of stored flours.

Experimental

In the first series of experiments 5 one-kilogram portions of flour were extracted for 3 days in a Schmidt (1916) ether extraction apparatus. The ether was removed from the extracted flour by spreading the flour in a thin layer and allowing the ether to evaporate at room temperature. The drying was allowed to continue for several days or until the flour gave no odor of ether when water was added to it. These flours and the data obtained in connection with them are listed in Table I. Flour No. 1 was a patent flour which had been stored in the laboratory for 5 years under

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very satisfactory conditions of storage, but the original pH of which was unknown. Flours 2, 3, 4 and 5 were a 3rd middling, a straight, a 1st clear and a 2nd clear, respectively, all freshly milled from the same wheat. Before the work reported here was done, however, the flours had aged for about a month. The ash contents, as given in Table I, will indicate the relative grade of the flours.

TABLE I

THE H-ION CONCENTRATION OF A STORED FLOUR AND OF SEVERAL FRESHLY MILLED FLOURS BEFORE AND AFTER EXTRACTION OF THE FLOURS WITH ETHER. THE QUANTITIES OF ACID EXTRACTED BY THE ETHER ARE ALSO GIVEN

Description of Flour	Ash (dry basis)	pH		Acid extracted in terms of cc. 0.1N acid per kilogram of flour
		Natural Flour	Extracted Flour	
	%			
Old patent	0.50	5.56	5.90	171.0
Freshly milled 3rd middlings	0.42	5.84	5.92	16.9
Freshly milled straight	0.61	6.09	6.23	25.7
Freshly milled 1st clear	0.90	6.22	6.33	59.8
Freshly milled 2nd clear	1.58	6.29	6.45	67.9

The H-ion concentration before and after extraction with ether and quantity of acid in the ether extract were determined. The extracts used for the H-ion concentrations were prepared by the method of Bailey and Johnson (1922), 10 g. of flour being suspended in 50 cc. of distilled water and after digestion for 1 hour at 25° C. the clear extract obtained by centrifuging. The H-ion determinations were made electrometrically using a Bailey (1920) hydrogen electrode. One-tenth N alcoholic KOH was used for the titration of the ether extract, with phenolphthalein as the indicator. The data in Table I show that extraction of these flours with ether reduced the H-ion concentration of the water extracts. The reduction was greatest in the case of the old patent flour, being from pH 5.56 to 5.90, which is equivalent to saying that the H-ion concentration changed from that of an old flour (patent grade) to that of a freshly milled one. It was this observation that suggested to the writers that the entire change in H-ion concentration which occurs in flours during aging might be due to ether-soluble acids, in which case the character of the acid-reacting substances could readily be determined.

The quantities of acid extracted from the the same flours are given in the table. It is to be noted that by far the largest quantity of acid was extracted from the old patent flour. For the other flours the quantity of acid extracted varied with the flour grade,

the 3rd middling flour yielding the least, and the 2nd clear yielding the most acid. Thus the acid extracted from 1 kilogram of the old patent, 3rd middling, straight, 1st clear and 2nd clear was equivalent to 171.0, 16.9, 25.7, 59.8 and 67.9 cc. of 0.1 N acid, respectively.

An effort was made to ascertain the nature of the acids extracted by determining the ratio of the distribution of the acids between ether and water. Portions of the ether extracts obtained in the preceding experiments were therefore concentrated, and to 100 cc. portions of the concentrate in tall narrow cylinders were added 100 cc. portions of water. After thorough shaking at frequent intervals during the course of several hours the cylinders were set aside to allow the separation of the water and ether into their respective layers. Fifty cc. portions of the water and ether layers respectively were then taken for titration with standard alkali. If most of the acids were found in the water layer this would indicate that the acids were of the lower fatty acid type, such as formic, acetic, etc., or hydroxy acids, such as lactic. If, on the other hand, most of the acids were found in the ether layer it would indicate that the acids were of the long carbon chain type aproaching palmitic and stearic. The coefficient of distribution or the ratio of the acid in one solvent to that in the other serves as a convenient way of expressing this relationship. The data obtained from the use of the ether extract from the old patent flour (Table I) will be given as characteristic of all the data. Titration of 50 cc. from the water and ether layers required 0.70 and 48.6 cc. of 0.1N alkali respectively. The ratio 0.70/48.6, hereafter called W/E, is therefore 0.014. Now since W/E for lactic acid varies between 9.4 and 12.2 depending on the concentration of acid used (International Critical Tables) it is obvious that the ether extract from flour can contain only small quantities of lactic acid, if it contains any lactic acid at all. The W/E values for formic, acetic, propionic, butyric and valeric acids decrease in general as follows: 2.50, 1.98, 1.52, 0.193 and 0.0734. The W/E values for the higher fatty acids in this series are not given in International Critical Tables. However, it is clear from the data given that on the average the fatty acids extracted from wheat flour with ether must have rather long carbon chains. A more exact study of the nature of the fatty acids in wheat flour will have to be left for future work.

In order to determine further whether an increasing content of fatty acids alone was responsible for the increasing H-ion concentration of flours during aging, there was obtained an old series of flours whose pH changes during aging were known. These flours

were made available to the writers through the courtesy of Dr. C. H. Bailey of the Minnesota Agricultural Experiment Station. The flours are described by Bailey and Johnson (1924) and the changes in H-ion concentration undergone by the flours are recorded in Table III of the Bailey and Johnson paper. On October 30, 1929, about 8 years after milling, these flours were subjected to extraction with ether and the quantity of ether-extractable acids, as well as the H-ion concentration, determined before and after extraction.

The water extracts used for the H-ion determinations were prepared in the manner already indicated. The ether was removed from the ether-extracted flour by spreading the ether-soaked flour

TABLE II.

THE H-ION CONCENTRATION OF A SERIES OF FRESHLY MILLED FLOURS, OF THE SAME SERIES AFTER STORAGE FOR 8 YEARS, AND AFTER STORAGE FOR 8 YEARS FOLLOWED BY ETHER EXTRACTION OF THE FLOUR

Chlorine per Barrel of Flour	H-ion Concentration of Freshly Milled Flour. (Nov. 9, 1921)	H-ion Concentration of Flours after Storage for 8 Years. (Oct. 30, 1929)	H-ion Concentration of Flours after Storage for 8 Years Followed by Ether Extraction	Acid Extracted from the Stored Flours in Terms of 0.1N Acid per Kilogram of Flour
Ounces	pH	pH	pH	cc.
0.00	6.04	5.60	5.97	261
0.42	5.93	5.55	5.87	233
0.50	5.90	5.62	5.92	255
0.58	5.88	5.35	5.63	284
0.67	5.87	5.35	5.51	280
0.75	5.83	5.55	5.85	220
0.83	5.80	5.58	5.95	275

in a thin layer and allowing the ether to evaporate. The data obtained are given in Table II. These data show that in some cases extraction of the 8-year-old flour with ether reduced the H-ion concentration to practically that of the freshly milled flour. Thus the original pH of the natural flour was 6.04; after 8 years aging it became 5.60; and after 8 years aging and extraction with ether it become 5.97. In the case of the flour bleached with 0.83 ounces of chlorine per barrel, the H-ion concentration of the old extracted flour was actually lower than that of the freshly-milled flour. No significance is attached to this, however, as the difference may be due to experimental error, especially as determinations of the H-ion concentration on this flour as reported by Bailey and Johnson (1924) were lower after 2 or 3 months storage than on the fresh flour. Thus the pH of this flour, 2 to 3 months old was 5.87; after 8 years storage, 5.58; and after 8 years storage and ether extraction, 5.95.

There were, however, two flours which did not return to their original H-ion concentration after extraction with ether; namely, those bleached with 0.58 and 0.67 oz. of chlorine per barrel, respectively. It was noted that the first of these had become infested with weevil during storage, which fact probably accounts for its different behavior. No weevils were found in the second flour, but as the flours were set aside for periods as long as 18 months without being looked at it would have been easy for weevil or some other agency to be responsible for its different behavior. It is also to be noted that these two flours yielded water extracts of significantly higher H-ion concentration than any of the other flours in this series.

Since ether extraction of old flours yielded, in most cases, flours of practically the same H-ion concentration as those of the fresh flours, it is believed that the increase in fatty acids produced during the storage of flour is responsible for the increase in H-ion concentration.

An additional series of flours was available which furnished data in support of this view. This series consisted of a patent, a 1st clear and a 2nd clear, all milled from the same wheat. This series of flours was obtained in August, 1926, and a large portion of each flour immediately extracted with ether. Again in November, 1929 portions of the unextracted flours were extracted with ether. The quantity of acid extracted with the ether was not determined in 1926 but was determined in 1929. The data which were obtained are given in Table III. These data indicate very clearly that the fatty acids formed during the storage of flour may alone be responsible for the increase in H-ion concentration which occurs during storage. Thus, the pH of the freshly milled patent flour was 5.93 (after ether extraction, 5.95); after 3 years storage the pH fell to 5.15; and after storage and extraction with ether the pH became 5.88, or practically what it was before storage of the flour. For the 1st and 2nd clear flours a similar series of changes in pH occurred on comparable treatment. Thus the original pH of the fresh 2nd clear was 6.33 (after ether extraction, 6.46); and after 3 years storage the pH fell to 6.12; and after storage followed by ether extraction it became 6.48, or almost identical with the pH of the fresh flour, ether extracted. During storage for 3 years the H-ion concentration of the flours which had been extracted with ether when fresh did not change. This is in agreement with what Bailey and Johnson (1924) had already observed. There was a decrease in H-ion concentration of the freshly milled

flours as a result of ether extraction, probably because they already contained some fatty acid extractable by the ether. As might be expected, the change in H-ion concentration due to ether extraction of the fresh flour varied with the fat content of the flours. Thus the data in Table III show that on the extraction of a patent, a 1st clear and a 2nd clear the changes in H-ion concentration were equivalent to 0.02, 0.07 and 0.13 pH units.

In Tables I, II and III there have been recorded the quantities of acid extracted with ether. According to Table I the quantities of acid extracted from fresh flours are relatively small. Thus from a 3rd middlings, a straight, a 1st clear, and a 2nd clear there was

TABLE III

THE H-ION CONCENTRATION OF FLOURS OF DIFFERENT GRADES BEFORE AND AFTER STORAGE FOR 3 YEARS AND BEFORE AND AFTER ETHER EXTRACTION. THE ETHER-EXTRACTABLE ACIDS ARE ALSO GIVEN

Description of Flour	Ash	H-ion Concentration of Freshly Milled Flours (August, 1926)		H-ion Concentration of Same Flours after Storage for 3 Years (Nov., 1929)		H-ion Concentration of 3 Year Old Natural Flour after Ether Extraction	Acid Extracted from the Stored Flour in Terms of 0.1N Acid per Kilogram of Flour
		Natural Flour	Ether Extracted Flour	Natural Flour	Ether Extracted Flour		
	%	pH	pH	pH	pH	pH	cc.
Patent	0.44	5.93	5.95	5.15	5.99	5.88	215
1st Clear	1.14	6.27	6.34	5.80	6.37	6.37	527
2nd Clear	1.70	6.33	6.46	6.12	6.48	6.48	1052

extracted from 1 kilogram of flour acid equivalent to 16.9, 25.7, 59.8 and 67.9 cc. of 0.1N alkali, respectively. During storage a marked increase in the ether-soluble acid occurs. Thus the data in Table III show that after storage for 3 years a patent, a 1st clear, and a 2nd clear flour yielded acid equivalent to 215, 527 and 1052 cc. of 0.1N alkali, respectively. It can be safely assumed that when the flours in Table III were fresh they contained ether-extractable acids of about the same quantity as the corresponding flours in Table I.

Collatz (1929) and Brooke (1929) have called attention to the many methods which have been used to determine the acidity of flour. The method most commonly used in the United States was the so-called tentative A. O. A. C. method in which the alkali-neutralizing substances in flours were extracted with water. In Europe the Balland or official Greek method was usually used, in which the extraction was conducted with 85% aqueous alcohol. Fifield and Bailey (1929) have made a comparison of the results

obtained when the two methods were used for the determination of the acidities of flours undergoing storage under various conditions. They concluded that the ratio of percentage of acidity, Greek method, to the percentage of acidity, A. O. A. C. method, was not constant through the range studied, but tended to become narrower as the percentage of acidity increased. It is to be expected that extraction with water in accordance with the A. O. A. C. method would, for the greater part, remove acid phosphates, while extraction with 85% alcohol would probably remove some phosphate salts in addition to organic fatty acids soluble in alcohol. In order to determine the nature of the acid-reacting materials present in the water extract (A. O. A. C. method), and in the alcohol extract (Greek method), the further experimental work reported in this paper was done.

TABLE IV

THE ACIDITY OF SEVERAL FLOURS OF DIFFERENT FLOUR GRADES AS DETERMINED BY DIFFERENT METHODS OF MEASUREMENT

Flour Grade	pH Value	Method of Measuring Acidity		
		A.O.A.C.	Greek	Ether Extraction
		cc. 0.1N acid per kilogram of flour	cc. 0.1N acid per kilogram of flour	cc. 0.1N acid per kilogram of flour
3rd middling	5.85	110	95	32
Straight	6.12	174	123	52
Straight bleached with chlorine	5.98	190	125	40
1st clear	6.20	279	189	90
2nd clear	6.27	466	335	198
Ground wheat	6.39	436	176	34

The acidities of a series of freshly-milled flours of different flour grades were first determined by the several different methods. The results which were obtained are given in Table IV. The acid-reacting substance extracted by the ether was much less than that extracted by either water or alcohol. Water extracted the largest quantity of acid-reacting substance, the water extract of the 3rd middling flour containing about 15% more acid than the alcohol extract of this flour. For the other flours the water extract contained about 40% more acid-reacting substances than the alcohol extract. The acidity of the ground wheat as determined by extraction with ether and with alcohol was rather low as compared with the acidity of the water extract. It is possible that the wheat was not ground finely enough to permit efficient extraction with ether or with 85% alcohol.

The extraction of the flour with ether was conducted by continually extracting for 72 hours, hence it may be assumed that

practically all of the organic acids of the flour have been removed. The fact that the acidity by the Greek method was so much higher than the acidity by the ether extraction method indicates that extraction with 85% alcohol by the Greek method must have removed a considerable quantity of material other than the usual organic acids. In order to study this further, several flours, which had been stored for some time and consequently were high in their ether-extractable acids, were subjected to extraction with ether and the acidity of the ether-extracted flours determined according to the A. O. A. C. and Greek methods. The results obtained are given in Table V. The data in this table are of interest in several

TABLE V

THE ACIDITY DETERMINED BY VARIOUS METHODS OF NATURAL FLOURS AND OF THE SAME FLOURS AFTER EXTRACTION WITH ETHER. THE FLOUR USED HAD BEEN SUBJECTED TO STORAGE FOR SOME TIME

Description of Flour	pH Value	Method of Measuring Acidity		
		A.O.A.C.	Greek	Ether Extraction
		cc. 0.1N acid per kilogram of flour	cc. 0.1N acid per kilogram of flour	cc. 0.1N acid per kilogram of flour
Natural Flour				
Straight flour No. 1	5.60	321	411	285
Patent flour No. 2	5.39	219	415	209
2nd clear flour No. 3	6.12	691	1240	1052
Patent flour No. 4	5.19	257	...	175
Ether Extracted Flour				
Straight flour No. 1	5.97	341	192	...
Patent flour No. 2	5.96	246	181	...
2nd clear flour No. 3	6.48	529	159	...
Patent flour No. 4	5.95	288	170	...

respects. First, except for the 2nd clear flour, the acidity, A. O. A. C. method, was higher after extraction with ether than before. This is no doubt due to the effect of ether extraction in producing a flour with much finer particles to which Johnson (1928) has already called attention. The finer flour is more readily and more completely extracted with water, hence the higher acidity of the water extract. However, some of the ether-soluble acid in the flour undoubtedly also dissolves in the water during extraction by the A. O. A. C. method and when the flour contains enough of the ether-soluble acid, sufficient of it may dissolve in the water to counterbalance the higher acidity which should result for ether extracted flours, in which case the flour has been brought to such a condition as regards particle size that more complete extraction (hence higher acidity) is possible. It is believed that this is the

explanation for the decrease in acidity effected by ether extraction of flour No. 3, in which case the ether-extractable acid per kilogram of flour was equivalent to 1052 cc. of 0.1 N NaOH, while for the flours which increased in their acidity (A. O. A. C.) on ether extraction, the ether-extractable acid was equivalent to only 200 to 300 cc. of 0.1N NaOH per kilogram of flour. It is obvious, therefore, that the acidity, A. O. A. C. method, of ether-extracted flour is the resultant of two phenomena operating in opposite directions. Whether ether extraction operates to increase or decrease the acidity of the water extract of the extracted flour as compared with that of the unextracted flour depends on which of the phenomena is dominant.

On comparing the acidities (Greek method) of ether-extracted and unextracted flour it appears that ether extraction removed a large quantity of the acid which would have been removed by the 85% alcohol. Thus, for the straight grade flour, No. 1, the acidity (Greek method) fell from 411 to 192 cc. of 0.1N NaOH on ether extraction, while for the 2nd clear flour, No. 3, the decrease was from 1240 to 159 cc. of 0.1N NaOH. The fact that the acidities (Greek method) of the ether-extracted flours were so nearly the same in spite of wide differences in ash content and acidities (all methods) of the unextracted flours tends to indicate that the 85% alcohol had dissolved to the saturation point other acid-reacting substances present in the extracted flours. These substances were probably the flour salts and flour proteins. It was noted that when ether was added to the alcohol extract a precipitate formed which had the appearance and properties of gliadin.

Now it is possible that the acidity by the Greek method could consist of the acidity, by the ether extraction method, plus the acidity of the ether-extracted flour, as determined by the Greek method. Data are available for verifying this opinion. Thus the calculated and determined acidities for flours Nos. 1, 2 and 3 in Table V are 477 and 411, 390 and 415, and 1211 and 1240, respectively. The agreement between calculated and determined acidity is not especially good, but it is close enough to warrant the suggestion that the acidity as determined by extraction with 85% alcohol is made up of 100% of the ether-soluble acidity plus another type of acidity not soluble in ether, but soluble in 85% alcohol. It is quite likely that the second part of the acidity is due in part to some of the substances which are responsible for the acidity of water extracts.

It has been shown earlier in this paper that the H-ion con-

centration of the water extract of an ether-extracted flour was practically the same as that of the freshly-milled flour, which indicated that the substances responsible for the increased H-ion concentration of old flours were fatty acids and consequently were removed by extraction with ether. Since the acidity, as determined by the Greek method, is influenced by the ether-extractable acids, it is possible that the increase in acidity (as measured by this method) which occurs when flours are aged is due entirely to the increase in ether-soluble acids. We have data which show that ether-soluble acids increase very rapidly during the storage of flour. Thus during storage at room temperature for 90 days a 3rd middling, a straight, a 1st clear, and a 2nd clear showed the following increases in ether-extractable acidity: from 32 to 73, from 52 to 125, from 90 to 236, and from 198 to 572 cc. of 0.1N NaOH per 1000 g. of flour, respectively.

If it is true that the ether-extractable acids determine the magnitude of the acidity as determined by the Greek method, then they should have relatively slight effects on the acidity determined by the A. O. A. C. method, since the ether-soluble acids would be relatively insoluble in water. The data of Fifield and Bailey (1929) support this view. Thus, for the first durum flour for which they give data, the acidity by the Greek method was 0.057. After 190 days storage at 25° C. the acidity became 0.167, i.e., increased by about 200%. For the same flour stored under the same conditions the acidity as determined by the A. O. A. C. method increased only about 66%. This indicates that the greater part of the acid material which developed in flour during storage was not soluble in water. Further evidence for this is furnished by certain data of Halton and Fisher (1928). Halton and Fisher stated that the H-ion concentration of a flour-in-water suspension was higher than that of the filtered or centrifuged extract. They further showed that when flour was allowed to settle from a flour-water suspension the H-ion concentration of the precipitate increased as the time the suspension was allowed to stand increased; i.e., as the volume occupied by the precipitate decreased. On the other hand, the H-ion concentration of the supernatant liquid decreased with time of standing. Our explanation for these observations is that when the flour suspension was filtered or the flour allowed to settle, the more acid-reacting, ether-soluble acids went with the flour, hence the filtrate or decantate would have lower H-ion concentration than the rich-in-flour phase or the precipitate, respectively. It may be argued that the fatty acids, probably being lighter than water,

should float, or at least be in the supernatant liquid. However, the fatty acids are probably absorbed on the heavier starch and protein particles of the flour, hence precipitate with them.

The experiments showing the ratio of distribution of the flour fatty acids between ether and water indicate that these acids have some slight solubility in water and hence could influence both the H-ion concentration and titratable acidity of the water extract. The effect of the ether-soluble acids on the titer of water extracts was evident only for the 2nd clear flour, in which case the titer of the water extract before ether extraction was equivalent to 691 and after ether extraction to 529 cc. of 0.1N NaOH per kilogram of flour. This decrease in titer was effected, however, by the extraction of ether-soluble acid equivalent to 1052 cc. of 0.1N alkali.

The data which have been presented indicate that the ether-soluble acids present in flour may alone be responsible for the changes in the acidity of the flour which have occurred during storage. The change in acidity may be demonstrated and measured in several ways, by change in H-ion concentration of the water extract as did Bailey and Johnson (1924), by change in acidity of the water or alcohol extract as did Fifield and Bailey (1929), or by changes in acidity of the ether extract. Since it is the changes in ether-soluble acids which are believed to be alone responsible for the changes in acidity as measured by any other method, it is obvious that the most direct method to determine the effect of aging or storage on the acidity of flour would be a determination of the ether-soluble acids. Further work on this phase of the problem has already been begun.

Summary

The H-ion concentrations of water extracts of stored flours extracted with ether were the same as those of the extracts of the freshly-milled flours.

The idea is expressed that ether-extractable acids are alone responsible for the changes in H-ion concentration or in acidity (as measured by any method) which occur in flours during storage under proper conditions.

The acids removed from flour by extraction with ether must, on the average, have rather long carbon chains as their coefficient of distribution between water and ether was of the order 0.014 to 1.

Only relatively insignificant quantities of lactic acid can be present in flour.

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A STUDY OF SOME METHODS OF EXAMINING FLOUR, WITH SPECIAL REFERENCE TO THE EFFECTS OF HEAT. II. EFFECTS OF HEAT ON FLOUR ENZYMES [1]

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The enzymic activities, diastatic and proteolytic, have at various times been considered of fundamental importance in baking quality. Reference must be made to the work of Baker and Hulton (1908), Ford and Guthrie (1908), Swanson and Calvin (1913) and Stockham (1920) and finally Rumsey (1922) and Collatz (1922), who contributed largely to our knowledge of diastatic

[1] Condensed from a Thesis approved for the degree of Doctor of Philosophy in the University of London.

activity. Wood (1906-8) considered two factors to be essential for the explanation of baking strength, one of which controlled the size of the loaf and the other controlled the shape. The size of the loaf he considered as a function of the gas-producing power, which is determined by the diastatic activity. Kent-Jones (1926) precludes the gas production factor from his definition of strength, because he maintains that it is very largely under the immediate care of the baker, who can easily remedy any deficiency in this respect.

Kent-Jones again has shown that the enzymes which actually saccharify soluble starch are apparently very sensitive to heat, while the more important enzymes, those capable of rendering soluble the normally insoluble starch, are very resistant to heat. He examined the diastatic activity before and after heating and found that correct heat treatment (i.e., for maximum baking strength) had very little effect either on the maltose content of the flour (Rumsey's autolytic method) or on the actual gas production of the flour. Severe heat treatment, however, was found to depress the gas production power of the flour. Kent-Jones, further, studied the Lintner values before and after heating; he showed that this value decreased on heating, the difference becoming progressively greater as the severity of the heating increased. The observations of the author have confirmed these findings. The same worker used the fall in viscosity of flour suspensions kept at 27° C. to indicate the proteolytic activity of the flour, and concluded therefrom that the proteolytic activity is not proportional to weakness, for some strong flours have quite marked activities, while many weak flours are lacking in this.

Berliner and Rüter (1928a) found that the heating of flour caused the water extracts to lose the capacity of splitting up a phosphatide which was obtained from a soya bean suspension, that is, the phosphatase activity is decreased by heating; this activity is said not to be affected by high temperatures if the water is first removed from the flour at a low temperature. It was found that the amount of water-soluble substances and the refraction number were diminished by heating, and that an increase in the H-ion concentration was accompanied by a decreased titratable acidity, which showed that the acidity change was due not to an increase in primary phosphate, but to a preponderating decrease in the secondary body. Heating also brought about a decrease in the conductivity of the aqueous extract of the flour as well as a decrease in the formol titration after Sørensen, which is indicative

that the hydrolytic splitting up of the flour proteins is prevented by heat.

Experimental

The particulars of the materials used in the present work have been given in the first part of this paper.

From the viewpoint of the present paper, the following enzymes are of importance and interest: diastase, phosphatase, protease and catalase. Others known to be present are cytase and peroxidase, but these will not be considered.

Phosphatase.—From a comparison of the changes in conductivity of flour extracts and those of phytin solutions in contact with the enzyme, Collatz and Bailey (1921) concluded that the conductivity of the water extracts of flour is chiefly due to the presence in solution of dissociated inorganic salts of phosphoric acid, which are produced by the hydrolysis of the phytin by the phytase present in the flour suspension.

As previously stated, Berliner and Rüter used soya bean as the source of the organic phosphate and the author decided to investigate the phosphatase activity with the flours of this study. In the same way, the soya bean was adopted in the first place as the source of the phosphatide and the extracts were prepared as follows:

Soya bean suspensions.

> The beans were finely ground in a coffee mill and 4 g. of this fine product suspended in 200 cc. of conductivity water; mixture was effected by vigorous shaking.

Flour extracts.

> 10 g. of flour was mixed with 100 cc. of conductivity water and shaken mechanically for 10 minutes; the suspension then stood for 20 minutes, when it was filtered through a No. 5 Whatman filter paper.

The method then followed was as given:

> To the 200 cc. of soya bean suspension was added 50 cc. of flour extract and the whole thoroughly mixed; the time from starting the extraction of the flour to the addition of the extract to the soya bean was uniform in all cases as far as possible. About 20 cc. of mixture was filtered immediately and the remainder placed in a closed flask in a water bath maintained at 37° C.; at the end of definite periods, portions were filtered off and the conductivity determined at 25° C. The conductivity set used was a ten-meter bridge set supplied by the Cambridge Instrument

Company, and satisfactory duplicates were obtained with it.

The results obtained are given in Table I.

TABLE I

PHOSPHATASE ACTIVITY AS SHOWN BY CONDUCTIVITY MEASUREMENTS OF A SOYA-BEAN FLOUR EXTRACT, WHEN INCUBATED AT 37°C.

Time	Flour I	Flour II	Flour III	Flour IV	Flour V	Flour VI
	Specific Conductivity x 10^{-4}.					
0 hrs.	7.20	7.20	7.86	7.74	7.74	7.64
2 "	9.14	7.86	8.06	8.05	8.05	8.05
15 "	11.25	12.00	11.47	10.52	11.32	11.00
24 "	11.61	12.67	12.40	11.54	12.40	12.00
39 "	12.49	13.95	13.23	12.72	12.90	12.20
47 "	14.29	14.51	14.29	13.80	14.25	14.08

The original specific conductivities of the mixtures were much higher than those reported by Berliner and Rüter, and there did not appear to be any appreciable diminution of enzymic activity due to the heating, even in the case of the strongly overheated flour No. VI. The reason for this difference from the work of Berliner and Rüter is not clear, but it must be remembered that the flours examined by them which showed this decreased activity, had been heated for 15 hours at 80° C. and for 15 hours at 90° C., which is over 50% greater than in flour No. VI. It may be that the point at which the enzymic activity is inhibited had not been reached even in the case of No. VI. Since this is the commercial article, the interest of the author is mainly in the changes occurring in that particular product, and experiments were not continued on flours which had been submitted to more severe treatment.

A number of individual wheat flours were similarly examined for phosphatase activity by the method of Berliner and Rüter, but only minor differences in the enzymic activity were found. A slightly greater phosphatase activity was found in a lower grade (ash 1.10%) than in a patent (ash 0.38%) flour from the same blend. Thus in 64 hours, the specific conductivity of the patent flour increased from 7.79×10^{-4} to 14.28×10^{-4} and that of the lower grade from 9.08×10^{-4} to 16.56×10^{-4}.

Another experiment was then undertaken, similar to the foregoing, but without the presence of the soya-bean suspension. That is, the flour-water suspensions were incubated at 37° C. and the conductivities of the extracts were determined periodically. In this

experiment, therefore, the sole source of organic phosphorus was the flour itself.

> 12.5 gm. of flour was added to 250 cc. of conductivity water and the whole well shaken to effect admixture. A portion was immediately filtered off for the zero measurement and the remainder placed in a closed flask in a water bath, maintained at 37° C; samples were then taken at the prescribed intervals and specific conductivities at 25° C. determined.

The results are given in Table II. The conclusions were the same as before, namely, that there was no appreciable difference between the heated and unheated samples. The rate of increase showed very definite decline as the time progressed, and in some cases an actual retrogression was noticed.

TABLE II

CHANGES IN SPECIFIC CONDUCTIVITY OF FLOUR-WATER SUSPENSIONS WHEN INCUBATED AT 37°C.

Time	Flour I	Flour II	Flour III	Flour IV	Flour V	Flour VI
			Specific Conductivity x 10^{-4}.			
0 hrs.	2.80	2.45	2.45	2.55	2.55	2.45
2 "	3.31	2.92	2.95	3.10	3.06	2.90
15 "	3.43	3.55	3.55	3.65	3.60	4.20
25 "	4.63	5.20	5.30	5.43	5.37	4.88
40 "	6.29	5.80	5.80	5.86	6.10	5.80
48 "	5.90	5.60	5.50	6.27	...	5.97
63 "	...	...	...	6.28	...	6.00

In as far, therefore, as heating was carried in these experiments, the effect on phosphatase activity is negligible. This was confirmed by determining the increased P_2O_5 contents of flour extracts after incubation at 27° C.; increases in six hours were 0.077% in the untreated and 0.071% in the overheated. The original P_2O_5 content was lower in the overheated flour.

Protease.—The importance of the proteolytic or proteoclastic enzymes has been emphasized by a number of workers. Ford and Guthrie (1908) used a gelatin liquefaction test, but regarded it only as a qualitative one. Stockham (1920) also used this, but Sharp and Elmer (1924) autolysed flour and studied the protein distribution periodically during the autolysis. Swanson and Tague (1917) used the now well-known formol titration of Sørensen for the determination of amino acids; Collatz (1922) employed the decrease in viscosity of a flour suspension as a measure of the proteoclastic activity of added malt extracts. Olsen and Bailey

(1925) studied the effect of the proteases of yeast in the manufacture of bread; they concluded that the proteases contributed by sound, normal, intact yeast cells (baker's yeast) are negligible in their effect upon the properties of gluten during a four or five hour fermentation period.

Denham and Scott-Blair (1927) used the formol titration under conditions different from those of Swanson and Tague and came to different conclusions. They contend that Swanson and Tague's higher results at 37° C. than at 25° C. were not due to an increased extraction but to greater enzymic activity. Cairns and Bailey (1928) made a long investigation of the proteoclastic activity of wheat flour; they applied eight chemical methods for the measurement of this factor, as well as the viscometric method. They recommend that the Sørensen formol titration method is the best suited for the measurement of proteolysis in flour, and stated that the amount which occurs, when suspensions of high grade flour milled from sound wheat are digested for 48 hours at 37° C., is small when measured in terms of the amino nitrogen which appears in the digest.

Kent-Jones (1926) studied the gelatin test, the protein distribution after autolysis and the autolytic viscosity method; he found the gelatin test to be qualitative but scarcely quantitative and was not satisfied with the protein distribution determinations. He recommended that the autolytic viscosity method in 1/10,000 auramine gave the best indication of proteolysis. Berliner and Rüter (1928a) criticized this and suggested pH 3.0 lactic acid as a bactericidal solvent but came to conclusions similar to those of Kent-Jones, namely, that heating had an inhibiting effect on the working of the proteolytic enzymes. It has already been shown in Part I (Cereal Chem. **8**:1-23) that neither of these solutions is sufficient as a bacterial inhibitant and that the decrease in viscosities is due to the presence of bacteria; if an efficient inhibitant is used then the viscosity decrease is small. This would suggest that proteolytic enzymes in flour are of small magnitude, unless the development of bacteria is allowed.

In view of the probable importance of bacteria in the supply of proteolytic enzymes, it was thought that possibly the failure to get satisfactory results on the gelatin test may have been due to incomplete inhibition of the bacteria. The author therefore repeated this test, ensuring the complete inhibition of bacterial growth; in order to do this in the presence of 1% gelatin solution, it was found necessary to use a 5% solution of ammonium fluoride

for the dissolving of the gelatin. Even with these precautions, reliable and dependable results were not obtained.

Attention was then turned to the Sørensen formol titration method as previous workers had got satisfactory results with this. The figures obtained by Denham and Scott-Blair's (1927) modification were distinctly higher than those either of Swanson and Tague (1917) or Kent-Jones (1926). In this it is undoubtedly sound to reduce the extract to the pH of unionized amino-acids, but flour extracts contain acid phosphates and organic acids, and the author fails to see where allowance for these is made. He therefore prefers to use the conditions laid down by Swanson and Tague (1917). The main consideration in this is that the two end points chosen should be at the same or approximately the same pH. The author prefers the rose-red tint of the phenolphthalein to the faint blue colour of the thymolphthalein, particularly if the solutions are turbid, but to confirm the position of the end points, a number of electrometric titrations were carried out alongside the colorimetric titrations. This showed that the two colorimetric end points chosen by the author were approximately the same and in the region of pH 8.0 - 8.1.

The preliminary work showed very clearly that for reproducible results, the conditions of grinding and extraction had to be very carefully standardized; the following was used throughout the subsequent work on proteolytic activity of wheat or flour.

> Fifty grams of the flour or finely ground wheat is added to 500 cc. of water and 3 cc. toluene. Extraction is carried out at 37° C. for 20 minutes with frequent shaking (about every 5 minutes). The suspension is centrifuged and then filtered through a Büchner funnel, using a No. 5 Whatman filter paper. This filtered extract is then placed in a stoppered flask and maintained at 37° C. in a water bath. The first reading is taken 6 hours after the addition of the extracting water, and subsequent readings at stated intervals. For each reading two 10 cc. portions are pipetted out, to one of these 5 cc. neutralized formaldehyde solution (pH 8.0) is added and then allowed to stand for 15 rose-red color of phenolphthalein (this is called the 2nd minutes, when it is titrated against 0.01N NaOH to the titre). Meanwhile the other 10 cc. of extract is titrated against 0.01N NaOH direct to the first faint pink of phenolphthalein (this is called the 1st titre). In each case the amount of phenolphthalein solution added is the same, namely three drops of a 0.5% neutral alcoholic solution. To the incubating extract in the flask, 1 cc. of toluene is

added every 24 hours; this alteration in volume is not likely to affect the result appreciably if the quantities indicated above are used.

Tables III and IV show selected results on wheats and flours.

The amino-nitrogen contents of the wheat extracts increase as incubation at 37° C. proceeds; the results are plotted in Figs. 1 and 2. Straight lines are obtained, the slopes of which may, according to Denham and Scott-Blair, be a measure of the proteolytic activity.

The change in the amino-nitrogen content of the flour extracts is negligible in all cases over a period of 118 hours. This would agree with the results obtained by Kent-Jones (1926) using the formol titration; he found that the amino-nitrogen content of a flour suspension increased only from 0.025% to 0.052% in three weeks.

Table V shows the amino-nitrogen contents of the unheated and the heated flour extracts. Here again the proteolytic activity is negligible in all cases, but the original amino nitrogen is lower in the strongly overheated flour than in the untreated and correctly heated flours; this may be due to a reduced solubility.

A further experiment showed that the lower grade flours had slightly greater amino-acid production than patent flours and the presence of 0.2% yeast increased the rate of production, but this only became significant after 48-72 hours.

The detection of proteolytic activity from an estimation of amino-acid content assumes the complete breakdown of the molecule; proteolysis may be present sufficiently to alter the physical characteristics of the protein without effecting cleavage. The diminution of viscosity on incubation in the absence of bacteria is, however, of small magnitude.

Although heating may have some effect on the proteolytic enzymes of flour, the data in this paper would indicate that the extent of the proteolytic activity of flour in a sterile medium is so small as to be of little significance. It must be remembered, however, that commercially flour is never sterile and it is probable from viscometric results that the effect of heating is to increase the resistance of the flour proteins to any proteolytic enzymes in the dough.

Catalase.—This enzyme has the power to break down hydrogen peroxide, and it is usually determined by this means; it is said to be present in flour—at any rate oxygen is liberated from a mixture of a flour suspension and hydrogen peroxide. Wender and Lewin

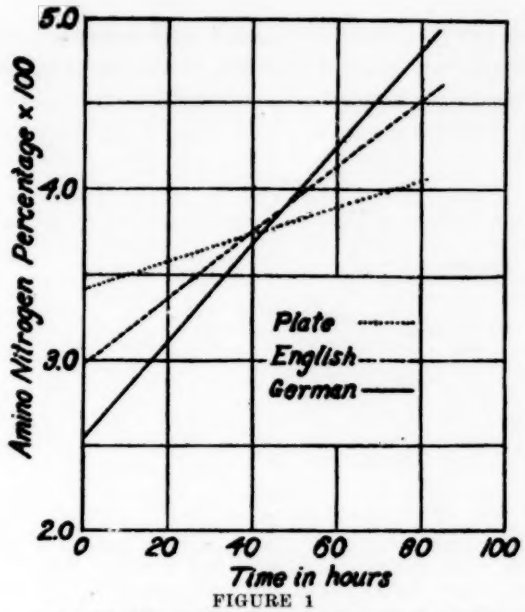

FIGURE 1

Amino-Nitrogen Content of Wheat Extracts During Incubation at 37° C. English, Plate and German Wheats are Shown

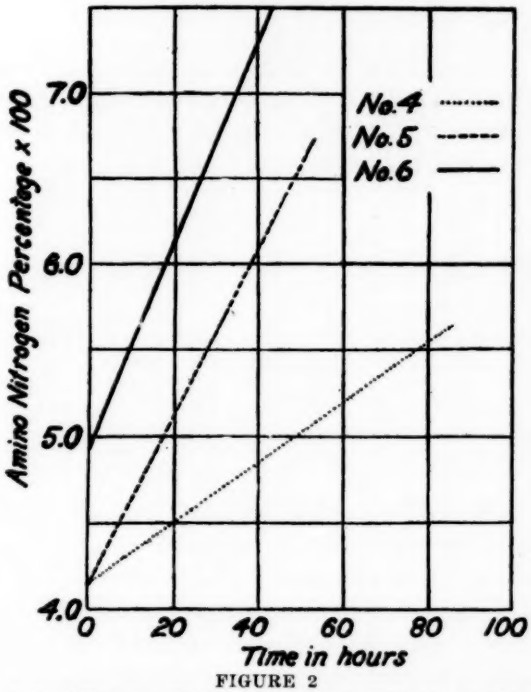

FIGURE 2

Amino-Nitrogen Content of Wheat Extracts During Incubation at 37° C. Nos. 4, 5 and 6 Manitoba Wheats are Shown

(1904), Miller (1909), Marion (1920) and Marotka and Kaminka (1922) have discussed the relationship of this to flour grade. Bailey (1917) described a convenient apparatus for the gasometric measurement of the oxygen evolved from a mixture of a flour sus-

TABLE III

AMINO-NITROGEN CONTENT OF WHEAT EXTRACTS DURING INCUBATION AT 37° C.

Time	English				Plate				No. 4 Manitoba			
	1st titre.	2nd titre.	Diff.	Amino N %	1st titre.	2nd titre.	Diff.	Amino N %	1st titre.	2nd titre.	Diff.	Amino N %
6 hrs.	2.7	4.9	2.2	0.031	4.1	6.6	2.5	0.035	4.1	7.3	3.0	0.042
12 "	2.7	5.0	2.3	0.032	4.1	6.6	2.5	0.035	4.1	7.3	3.2	0.045
24 "	2.7	5.2	2.5	0.035	4.1	6.7	2.6	0.036	4.1	7.4	3.3	0.046
36 "	2.7	5.3	2.6	0.037	4.1	6.7	2.6	0.036	4.1	7.5	3.4	0.048
48 "	2.7	5.5	2.8	0.039	4.1	6.7	2.6	0.036	4.1	7.5	3.4	0.048
60 "	2.7	5.6	2.9	0.040	4.1	6.8	2.7	0.039	4.1	7.8	3.7	0.052
	No. 5 Manitoba				No. 6 Manitoba				German			
6 hrs.	3.3	6.4	3.1	0.043	3.5	7.3	3.8	0.053	2.8	4.7	1.9	0.027
12 "	3.3	6.6	3.3	0.046	3.5	7.7	4.2	0.059	2.8	4.8	2.0	0.028
24 "	3.3	7.2	3.9	0.055	3.5	8.0	4.5	0.063	2.8	5.2	2.4	0.034
36 "	3.3	7.5	4.2	0.059	3.5	8.8	5.3	0.074	2.8	5.4	2.6	0.036
48 "	3.3	7.6	4.3	0.060	3.5	9.0	5.5	0.077	2.8	5.6	2.8	0.039
60 "	..	..	..	....	..	..	..	....	..	..	..	....

TABLE IV

AMINO-NITROGEN CONTENT OF FLOUR EXTRACTS DURING INCUBATION AT 37° C.

Time	Australian				Plate				No. 4 Manitoba			
	1st titre.	2nd titre.	Diff.	Amino N %	1st titre.	2nd titre.	Diff.	Amino N %	1st titre.	2nd titre.	Diff.	Amino N %
1 hr.	1.1	2.2	1.1	0.015	1.5	2.7	1.2	0.017	1.5	2.9	1.4	0.017
24 hrs.	1.0	2.1	1.1	0.015	1.5	2.7	1.2	0.017	1.5	2.8	1.3	0.019
48 "	0.9	2.0	1.1	0.015	1.5	2.8	1.3	0.019	1.5	2.8	1.3	0.019
72 "	0.9	2.0	1.1	0.015	1.5	2.7	1.2	0.017	1.5	2.8	1.3	0.019
96 "	0.9	2.0	1.1	0.015	1.5	2.7	1.2	0.017	1.4	2.7	1.3	0.019
118 "	0.8	1.9	1.1	0.015	1.5	2.8	1.3	0.019	1.3	2.6	1.3	0.019
	No. 3 Manitoba				No. 6 Manitoba				English			
1 hr.	1.3	2.7	1.4	0.020	1.6	3.5	1.9	0.027	1.0	2.1	1.1	0.015
24 hrs.	1.3	2.6	1.3	0.019	1.6	3.5	1.9	0.027	1.0	2.2	1.2	0.017
48 "	1.3	2.8	1.5	0.021	1.6	3.5	1.9	0.027	1.0	2.2	1.2	0.017
72 "	1.3	2.7	1.4	0.020	1.6	3.5	1.9	0.027	1.0	2.2	1.2	0.017
96 "	1.3	2.7	1.4	0.020	1.6	3.5	1.9	0.027	1.0	2.3	1.3	0.019
118 "	1.3	2.7	1.4	0.020	1.4	3.4	2.0	0.028	1.0	2.3	1.3	0.019

pension and hydrogen peroxide and this apparatus was used for submitting the heated and unheated flours to this test.

The significance of the catalase activity from a baking point of view is not known, but it was thought to be of interest to study the effect of heat upon this enzymic activity. Table VI gives the results; readings were taken in duplicate in all cases and they have been corrected to 0° C. and 760 mm.

It will be seen that the very strong heating of flour results in a reduced catalase activity, although there is nothing to show whether this is a solubility effect or strictly a lowered enzymic activity.

TABLE V

AMINO NITROGEN CONTENTS OF UNHEATED AND HEATED FLOUR EXTRACTS DURING INCUBATION AT 37°C.

Time	Flour I				Flour II				Flour III			
	1st titre.	2nd titre.	Diff.	Amino N %	1st titre.	2nd titre.	Diff.	Amino N %	1st titre.	2nd titre.	Diff.	Amino N %
1 hr.	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014	1.0	2.0	1.0	0.014
24 hrs.	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014	1.0	2.0	1.0	0.014
48 "	0.9	1.8	0.9	0.013	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014
72 "	0.9	1.8	0.9	0.013	0.9	1.9	1.0	0.014	1.0	1.9	0.9	0.013
96 "	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014	1.0	2.0	1.0	0.014
118 "	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014	1.0	2.0	1.0	0.014
	Flour IV				Flour V				Flour VI			
1 hr.	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014	0.7	1.5	0.8	0.011
24 "	0.9	1.9	1.0	0.014	0.9	1.9	1.0	0.014	0.7	1.5	0.8	0.011
48 "	1.0	1.9	0.9	0.013	0.9	1.9	1.0	0.014	0.7	1.5	0.8	0.011
72 "	1.0	1.9	0.9	0.013	0.9	1.8	0.9	0.013	0.7	1.5	0.8	0.011
96 "	1.0	1.9	0.9	0.013	0.9	1.9	1.0	0.014	0.7	1.5	0.8	0.011
118 "	1.0	1.9	0.9	0.013	0.9	1.9	1.0	0.014	0.7	1.5	0.8	0.011

Dough acidity changes during fermentation.—As mentioned in Part I of this paper, the changes in pH of a dough during fermentation are of small magnitude, but many chemists have believed that the acid produced during the fermentation of the yeast must play

TABLE VI

CATALASE ACTIVITY OF FLOUR SUSPENSIONS BEFORE AND AFTER HEATING

Flour	Cc. Oxygen Liberated During One Hour at 37°C.		
	(a)	(b)	Average
I (unheated)	3.1	2.7	2.9
II (heated)	3.2	3.0	3.1
III (heated)	2.6	3.0	2.8
IV (heated)	3.0	3.0	3.0
V (heated)	2.5	2.5	2.5
VI (strongly overheated)	0.85	0.75	0.80

some rôle in bread making. Bailey (1925) emphasized the importance of acidity on the working of the enzymes.

In the work which follows, the doughs were prepared normally with tap water and the baking procedure was the test baking method of Kent-Jones (1927, p. 174). The whole process is by

hand and a report is obtained at each stage of the fermentation; the dough is fermented for 1½ hours to the first punch and it is then "knocked" back and replaced in the proving cabinet for a further half hour; after this time it is rounded into balls and allowed to stand on a board (covered by a cloth) for 15 minutes. It is then moulded into its required shape and given the final "proof" of 30 to 40 minutes and then to the baking oven. Dough pieces were taken at each stage and the suspensions prepared using 15 g. of dough and 95 cc. of water; this was shaken for 10 minutes and filtered after 30 minutes. Upon these filtered extracts two observations were made:

(I) pH. Quinhydrone method
(II) titratable acidity. 10 cc. titrated against 0.01N alkali, using phenolphthalein as indicator.

In working with this strongly overheated flour improver some years ago, it was noticed that the addition of 0.1 cc. syrupy lactic acid to the doughing water of a loaf caused an increased oven spring, so included in these experiments were some in which flours were treated:

(a) with 0.7% strongly overheated flour improver.
(b) with 0.7% above improver plus 0.1 – 0.5 cc. syrupy lactic acid, added to the doughing water of a loaf (655 g. flour).

Typical results are given in Table VII.

TABLE VII

Acidity of Doughs During Fermentation. Straight Run Flour Used

Sample Taken	Untreated		Treated 0.7% Heated Flour		Treated 0.7% Heated Flour Plus Acid	
	pH	Titration cc. 0.01N NaOH per 10 cc.	pH	Titration cc.	pH	Titration cc.
Flour.	6.20	1.3	6.20	1.3	6.20	1.3
Make.	6.00	1.4	5.90	1.5	5.70	1.6
1st punch.	5.90	1.5	5.85	1.5	5.60	1.8
Moulding.	5.85	1.6	5.85	1.6	5.50	1.9
To oven.	5.85	1.7	5.85	1.6	5.50	2.0
Bread.	6.00	1.6	5.95	1.5	5.60	1.9

In the case given 0.5 cc. syrupy lactic acid had been used, so that the pH of the doughing water was 3.60. It was noticed throughout fermentation that this "acidified" dough had much greater liveliness and freeness and the oven spring was distinctly better. As can be seen, the pH of the dough was altered slightly, but with

smaller quantities of acid there appeared practically no alteration, although the modification of the dough properties was always present. It seems, therefore, that even though considerably more acid is added to the water, the buffering value of the flour is sufficient to prevent the pH of the suspension changing to any large extent. This raises an interesting question as to whether the pH of the dough suspension has any relation to the pH of the dough, and secondly, whether the pH of the aqueous dispersion medium in the dough is the same as the pH of the dough fibrils—it may be possible, by the addition of acid, to modify the pH of the aqueous phase and so accelerate enzymic activity therein, without affecting the acidity of the solid structure of the dough. Such a condition would be completely masked immediately the suspension of the whole dough is made. This hypothesis would account for the observed increased activity of the yeast and enzymes without any general effect upon the H-ion concentration of the dough mass.

A number of doughs were studied with a variety of chemical and physical treatments; there appeared to be two factors concerned in dough improvement: (a) increased stability, (b) increased spring and liveliness. The second may be due to an increased acid production, affecting the aqueous phase in the same way as the addition of acid to the doughing water. The over-treated flour improver possesses (a) as does potassium bromate to a marked degree, but ammonium persulphate carries out the joint action. It is suggested that the increased stability is concerned with the physical condition of the gluten fibrils and is probably a colloidal problem, whereas the increased spring and liveliness is connected with the acidity of the aqueous phase, and its effect on the enzymic activities.

Further work showed that doughs made with distilled (pH 6.00) water and rain water (pH 6.80) had rather more life and spring than a dough from the same flour made with ordinary tap water (pH 7.00), and greater oven spring was obtained. The differences in the acidity readings were negligible. The purer waters probably do not buffer the acidity changes in the aqueous phase to the same extent as the hard tap water.

Importance of Moisture on the Effects of Heat on the Enzymic Activity of Flour

It has already been seen (Part I) that if a large portion of the natural moisture in flour be removed, then heating produces the

characteristic alteration in the proteins at a greatly diminished rate. A similar examination was made of the effects of heat on the enzymic activities of a partially dried flour. As typical enzymes, which are sensitive to heat, it was decided to use the Lintner figure and the catalase activity.

(a) Catalase activity.

By Bailey's method, as previously referred to.

(b) Lintner degree.

Determined according to the procedure for flour, given by Kent-Jones (1927). The amounts of extract used in the present investigation were 0.1, 0.2, 0.5, 0.7, 1.0, 1.3, 1.5, 1.7, 2.0, and 2.5 cc. so as to give a somewhat wider range than that referred to. It must be remembered that this method is not designed for great accuracy.

TABLE VIII

ENZYMIC ACTIVITY, AS EVIDENCED BY CATALASE AND LINTNER FIGURES, AFTER HEATING FOR VARIOUS TIMES AT 180°F. IN THE PRESENCE OF MINIMUM MOISTURE

Time of Heating	Degrees Lintner	Cc. O_2 Evolved From One Gram Flour in 60 Minutes
Nil	30.0	3.0
11 Hours	12.0	0.8
22 "	12.0	0.7
46 "	9.0	0.7
112 "	2.0	0.7

The results obtained are given in Table VIII. On the evidence of this table, it would appear that enzymic activity is susceptible to heat even in the presence of minimum moisture content; thus after eleven hours' heating at 180°F., there is a definite decrease in the enzymic activity in both cases. The degree Lintner decreases further as heating is prolonged, but the catalase activity remains more or less unchanged.

The significant fact that enzymic activity is affected, but that protein changes are not at first produced, is strong evidence that the effects of this heated flour improver are dependent upon the altered conditions of the proteins, and not upon the enzymic activity.

Discussion

The heating of the whole of the flour so as to produce maximum baking strength does not affect enzymic activity to any appreciable extent and, therefore, it can be said quite definitely

that the improved behaviour of the dough is not due to any altered properties of the enzymes.

The strongly overheated flour has, however, been modified in these activities; some enzymes seem to be more sensitive to the effect of heat than others, but seeing that only 0.7 – 1.0% of this flour is used to give the desired effect, it can hardly be suggested that a diminished activity in this 1.0% is able to produce so pronounced an effect in the dough.

Berliner and Rüter have suggested that with strongly heated flour the absolute quantity of inorganic phosphates which arise in their aqueous suspension, decreases and the quantity of the secondary one more so than that of the primary one. Further, the same authors produced figures to show that the phosphatase activity of a flour was diminished by heating, indicating presumably that the quantity and nature of the phosphates present have some influence in determining the property of the heated flours. The present author is of the opinion that the changes in the acidity figures are too small to be of any significance in the explanation of the alteration of flour properties brought about by heating. The observations recorded here do not show any perceptible diminution in phosphatase activity, even after heating for 10 hours at 180°F.

The fact that flour, if previously dried at low temperatures, can be heated so as to diminish enzymic activity without acquiring any qualities of an "improver" indicates that this "improving" action is independent of enzymic activity. The usefulness of the overheated flour is, however, shown to be very closely related to the altered physical condition of the proteins.

The results on proteolytic enzymes are in accordance with previous workers in this field, in that in the absence of bacterial growth the activities are of small value. It is suggested that in the auramine method of Kent-Jones, the greater part of the proteolysis taking place in a flour-water incubation is due to the proteolytic enzymes produced by bacterial activity during the incubation. Another suggestion is that by overheating flour, the proteins are in some way brought to a condition in which they are acted upon only slightly, if at all, by proteolytic enzymes. This may correspond to a coagulating or hardening process. There is, perhaps, little evidence for this, but if it be assumed, it would possibly account for the anomalous behaviour of the strongly heated flour, for example, the effects of malt extracts on the heated flour, as noticed by Kent-Jones (1926).

It is further suggested that the results obtained by the auramine viscosity method are the measurement of the resistance of the proteins in flour to the proteolytic activity produced by the bacteria, rather than the measurement of the actual activities of the proteolytic enzymes of the flour itself.

Summary

The diminution in phosphatase activity, observed by Berliner and Rüter, was not confirmed in flour heated for 10 hours at 180°F. or less.

Proteoclastic activity of flour itself, as evidenced by amino nitrogen produced in an incubating extract, is of small value and can have little influence in a fermenting dough. The addition of 0.2 g. yeast to 100 cc. of extract increased the proteoclastic activity. Heating may increase the resistance of proteins to enzymic attack.

Catalase activity of flour suspensions is reduced by severe heating.

Confirmation is obtained that the change in H-ion concentration of a dough during fermentation is of small magnitude. It is suggested that the pH of the aqueous phase, however, may alter and affect the enzymic activity.

If a large proportion of the natural moisture of a flour be removed at a low temperature and then the heating performed, the enzymic activity appears to be inhibited at practically a normal rate.

Acknowledgment

The author desires to place on record his great appreciation of the continual interest which Doctor D. W. Kent-Jones has taken in these researches.

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EFFECT OF MOISTURE CONTENT OF FLOUR ON HEAT OF IMBIBITION DEVELOPED DURING THE MIXING OF BREAD DOUGH

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That the moisture content of cereals and cereal products varies according to the temperature and humidity of the surrounding medium, has been known for many years and has been the subject of study by many investigators. As early as 1883 Brewer found that the amount of moisture in wheat depended upon the dryness of the air.

Freshly milled flour has about 13.5% moisture. When it is held in heated rooms, it may lose one-third of this moisture as it establishes an equilibrium with atmospheric humidity. Bailey (1920) observed that flours exposed to different atmospheric conditions, the temperature in all cases being 25° C., ranged from 5% moisture at 30% relative humidity to 15% moisture at 80% relative humidity.

Daniels, Kepner and Murdick (1920) found the heat of hydration of various flours to range from 7.6 B.t.u. per pound in a low grade to 5.4 B.t.u. per pound in a winter-wheat flour.

In the bake-shop, 78-80° F. is regarded as the most favorable temperature for panary fermentation. In practice, allowance is made for heat developed during mixing, temperature of the room, and temperature of the flour. Ice or cold water is used to bring the dough to a definite temperature as it leaves the mixer. Referring to mixing temperature, Harrel (1926) says: "Temperature is equally as important as mixing time or revolutions per minute," and that temperature of the finished dough is one of the factors "which must constantly be kept in mind by the research worker and bread manufacturer." In recent years the American Association of Cereal Chemists has given considerable attention to the establishment of a standard baking test and to factors that cause variations in the finished loaf.

In a study made by this writer in 1930 in connection with the utilization of dairy products in baking, it was observed that the moisture content of the flour was a factor in the temperature of the dough as it was taken from the mixer. Because of the importance of the temperature of the finished dough, this study was made to demonstrate the effect of moisture content of flour on heat

evolved during mixing, and to call attention to the magnitude of its effect. It is not practicable to attempt to give data which can be reproduced quantitatively under conditions other than those of this experiment, because of variations in the baking practice, due to differences in type of mixers, differences in quantity of dough, and differences in length of time of mixing, etc.

Experimental

A 95% patent flour made from hard red winter wheat, containing 0.43% ash and 10.8% protein, was used in this study. When it reached the laboratory, a 2-quart jar was filled with some of the flour and the jar was sealed. In order to have flours of different moisture contents, three other samples of the same flour were held in the laboratory for different lengths of time and then placed in jars and sealed. The moisture contents of these four samples as used in the experiment are given in Table I.

TABLE I

Effect of Moisture Content on Heat of Imbibition of Bread Dough. Room and Ingredients at 28.5° C. at Beginning of Mixing Period

Moisture in Flour	Temperature of Dough
%	° C.
13.1	30.1
11.5	30.8
9.9	32.1
8.7	33.4

Heat of Imbibition of Bread Dough.—The dough was made acording to the formula in use by The American Association of Cereal Chemists and described by Harrel (1929).

Ingredients:
Flour, 100.0 g. on 15% moisture basis (85.0 g. dry matter)
Yeast, 3.0 g. (3%).
Salt, 1.0 g. (1%). 99.5% pure.
Sugar (sucrose), 2.5 g. (2½%).
Water (distilled), to 58% absorption with flour on a 58% moisture basis.

Portions equivalent to two loaves were used. The temperature of the room and each of the ingredients was 28.5° C. at the beginning of the mixing period. The dough was mixed three minutes in the three-quart bowl of a Hobart mixer with the hooks described by the Hobart Manufacturing Company as C-3-R and C-3-ER. The Hobart mixer was selected because of its low heat of friction. Four thermometers that could be read to 0.1° C. were placed in the dough at the end of the mixing period, and readings were made as the thermometers came to constant temperature. The average of these four readings was regarded as the temperature of the dough. Three trials were made on each flour and the average of the results

recorded in Table I. These data also are reproduced graphically in Fig. 1.

It will be observed that with decrease in moisture content of flour, there was an increase in the temperature of the finished dough. The change in temperature per unit change in moisture was greater in the flours with low moisture content than in those with the higher moisture content. Flour with a moisture content of

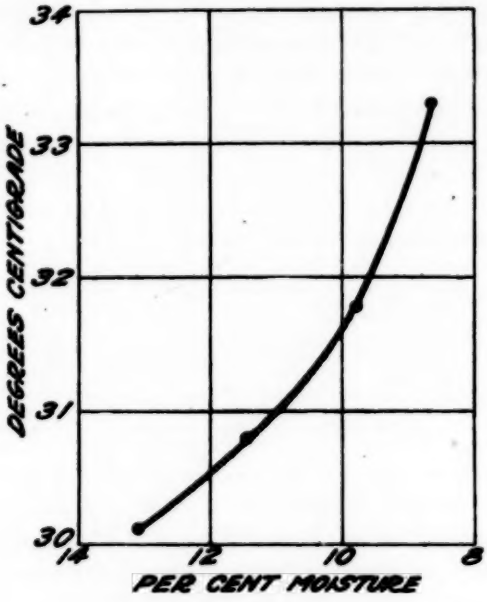

FIG. 1
Effect of Moisture Content of Flour on the Temperature of Bread Dough at the end of the Mixing Period.

13.1% produced a dough with a temperature of 30.1° C., whereas the flour with a moisture content of 8.7% produced a dough having a temperature of 33.4° C. The difference of 3.3° C. in temperature is of sufficient magnitude to be important in bread making.

When the values for moisture content are plotted against the logarithms of the variation in temperature (Figure 1) the resulting graph is practically a straight line, indicating that the variation in temperature developed with decrease in moisture content of flour is of a logarithmic nature.

Heat liberated during mixing is due partly to friction and partly to absorption of water by the flour. When conditions of this experiment are compared with bake-shop conditions, the differences in temperature changes noted are minimum rather than maximum. The loss of heat by radiation is large compared to that when large quantities of flour are used.

Summary

1. Heat of imbibition of suspensions and bread doughs varies with the moisture content of the flour.
2. Flours with moisture contents of 13.1% and 8.7% produced finished doughs which differed 3.3° C. in temperature, which is of sufficient magnitude to be important in bread making.

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A QUANTITATIVE MEASUREMENT OF THE CARBON DIOXIDE EVOLVED IN AND LOST FROM SIMPLIFIED MUFFIN BATTERS

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For many years experiments have been reported which show that some types of baking powders quickly release a large proportion of their carbon dioxide upon being mixed with an excess of water, while others evolve but little. No work, however, has been described which deals with the behavior of baking powders when combined with several ingredients into a batter or dough instead of with water alone, and yet this is one of the most significent points in the use of baking powders. A study of this question, therefore, was undertaken by measuring, (a) the quantity of carbon dioxide evolved in, and (b) the quantity lost from a batter or dough during its preparation. The present paper is a report of the progress made thus far.

For both series, a tartrate, a monocalcium phosphate, and two sodium aluminum sulfate-phosphate commercially prepared bak-

ing powders were used. These were combined with: (1) water alone; (2) a standard mixture consisting of 42.0% flour, 3.0% baking powder, 8.2% fat, and 46.8% water; (3) the standard mixture in which milk was substituted for water; and (4) the standard mixture to which sugar was added. The dry ingredients were mixed together and placed in a specially constructed mixing bowl into which extended an electrically driven paddle, an intake for the liquid ingredients, and an out-take for the gas. The temperature of all of the ingredients and also of the room was approximately 22° C.

TABLE I

CARBON DIOXIDE EVOLVED WHEN BAKING POWDER IS COMBINED WITH WATER ONLY, AND WITH OTHER INGREDIENTS INTO BATTERS

Type of Baking Powder	Average quantity of carbon dioxide evolved when baking powder is mixed for 40 seconds with:				
	Water	Flour, Fat, Water	Flour, Fat, Milk	Flour, Sugar, Fat, Water	
				4.9% Sugar	9.8% Sugar
	% by wt.	% by wt.	% by wt.	% by wt.	% by wt.
Tartrate	11.27	8.27	7.19	8.05	8.15
Phosphate	8.17	6.92	6.48	6.70	7.16
S.A.S.-Phos. No. 1	5.04	3.96	3.81	3.92	4.19
S.A.S.-Phos. No. 2	2.59	2.46	2.23	2.08	2.09

In the first series, which, as was indicated above, concerned the quantity of carbon dioxide liberated, the gas (or its equivalent volume of air) was collected in the gas burette of a Chittick carbon dioxide apparatus and measured as in the official A.O.A.C. gasometric method, by displacement of a saturated aqueous solution of sodium chloride, saturated also with carbon dioxide.

It was found, as is shown in Table I, that baking powder when combined with flour, fat, and water into a smooth batter evolved less carbon dioxide than when combined with water only. The difference was slight when the phosphate and the sodium aluminum sulfate-phosphate powders were used, but appreciable, about 25%, when the tartrate was used. It was found also that all the types of baking powders evolved slightly more carbon dioxide when water was used in preparing a batter than when an equal volume of milk was used. This result may be due, possibly, to the fact that there is less liquid present in the latter series, since milk is approximately only 87% water. The addition of sugar up to 9.8% of the ingredients was found to have no consistent effect upon the evolution of gas.

In the second series, which, as already mentioned, dealt with

the loss of carbon dioxide, the gas was swept out of the mixing chamber for one hour, dried in a sulfuric acid tower, and the carbon-dioxide fraction absorbed in weighed soda-lime tubes.

The results for this series are given in Table II. As may be seen from the table, it was found that the flour, fat, baking powder, and water mixtures lost between one-half and two-thirds as much carbon dioxide during their preparation[1] as did the corresponding baking powder and water mixture. The batters containing water lost slightly more carbon dioxide during their preparation than did similar ones containing an equal volume of milk. The addition of sugar up to 9.8% of the ingredients seemed to have no effect upon loss of carbon dioxide from a batter. A mixing period lasting just long enough to produce a smooth batter caused only slightly less loss of carbon dioxide than did a period twice this long.

TABLE II

CARBON DIOXIDE LOST WHEN BAKING POWDER IS COMBINED WITH WATER ONLY, AND WITH OTHER INGREDIENTS INTO BATTERS

Type of Baking Powder	Average quantity of carbon dioxide lost when baking powder is mixed for 40 seconds with:					Average quantity of carbon dioxide lost when baking powder is beaten for 20 seconds with flour, fat, and water
	Water	Flour, Fat, Water	Flour, Fat, Milk	Flour, Fat, Sugar and Water		
				4.9% Sugar	9.8% Sugar	
	% by wt.	% by wt.	% by wt.	% by wt.	% by wt.	% by wt.
Tartrate	10.11	6.77	6.01	6.45	6.78	6.18
Phosphate	9.34	5.56	...	4.71	5.81	4.52
S.A.S.-Phos. No. 1	5.16	3.25	2.78	2.58	2.60	2.70
S.A.S.-Phos. No. 2	3.91	2.29	1.76	1.89	1.61	1.27

A comparison of the results of these two series brings out the important point that during mixing a large proportion of the gas evolved is actually lost from the simplified batter mixtures. Thus, for example, when the tartrate or phosphate types were used, the batter lost approximately four-fifths of the gas evolved, and when the sodium aluminum sulfate-phosphate powder No. 1 was used about three-fourths or slightly less. With the other sodium aluminum sulphate-phosphate powder, however, the individual determinations overlapped so extensively that probably no general conclusion should be drawn from the averages until further work has been done. Whether similar losses are sustained by batters which contain egg and a greater proportion of sugar than these must be determined by subsequent experiment.

[1] The losses reported here are of course those sustained by the batter during the mixing period plus the one-hour collection period, as may be seen from the description of the method. An attempt to evaluate the loss during each of these periods is now under way.

TESTING NEW WHEAT VARIETIES

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(Read at the Convention, May, 1930)

New wheat varieties are in demand. In nearly all the wheat growing sections of North America there is an active interest in new varieties. Geneticists and agronomists in many of the experiment stations located in wheat areas, as well as federal workers, are devoting attention to the development of new varieties suitable for the widely different agronomic conditions of the many wheat-growing localities. Nowhere is the situation more acute than in the Northwest, where the ravages of rust epidemics present serious difficulties in growing spring wheat.

In this paper will be discussed certain aspects of wheat and flour testing which are applicable to wheats in general. While illustrations are chosen from recent tests of Northwest wheats, it is recognized that most of the problems of developing and testing new varieties are common to all wheat sections of the country.

It is not the bread bakers who are demanding new wheat varieties; in fact, it is probably a rare occurrence when a baker even knows the variety of wheat from which his flour is milled. Neither are the flour millers demanding new wheats, although it is obvious that most millers know the wheat varieties they purchase and grind. Millers are reasonably well satisfied with the better types of wheat which are now being grown and might be content with these varieties for many years, providing that the supply continued to be ample and of the same high quality.

The demand for new wheat varieties can be traced to the producers and there are good reasons supporting this demand. Millers are going to be affected greatly by the spread of new wheats and therefore the millers and chemists should be vitally interested.

Why are the wheat producers seeking new varieties? The answer is not difficult to find if we trace the trend in wheat production. Plant diseases are reducing yields per acre, and the quality of the grain to a level so low that wheat is no longer a profitable crop in many parts of the Northwest. Increases both in land value and annual carrying charges are additional factors which are pushing the wheat line westward and northward. Among plant diseases, rust, with over 100 known physiologic forms, takes the greatest toll. Smut, scab, root rot and black chaff are other diseases which complicate the problems of the wheat breeder. In a recent paper

Hayes (1929) has discussed the breeding of wheats for disease resistance. Winter hardiness is of prime importance in determining the value of fall-sown varieties, especially in the Northwest, where, as a result of the development of new varieties capable of withstanding severe cold, the winter wheat acreage has greatly increased in the last decade.

As cereal chemists cooperating with flour millers, we must direct our attention to milling and baking characteristics of new wheats. It cannot be disputed that the requirements of the producers of wheat must be met. Such requirements are becoming increasingly complicated.

What are some of the standards for an ideal wheat variety? First, large yield per acre, for this determines in large measure the farmers' income. In addition the wheat should be resistant to the multitude of plant diseases; if fall sown it should be cold resistant to withstand severe winters. It should withstand drouth; it should ripen early; it should have a stiff straw to prevent lodging when a wet season produces heavy growth; it should have long, well-filled heads which do not shatter when harvested, but which are easily threshed by machines without cracking the kernels; it should have uniformly plump kernels of dark, vitreous character to grade high with our present grading system; and, finally, it should have desirable milling and baking properties. It is doubtful whether a single variety of wheat can be found to meet all these requirements. The problems of the agronomist and plant breeder are complicated enough without the necessity of satisfying the cereal chemist who has set up high standards for new wheat varieties.

Several new wheat varieties have appeared in the last ten or fifteen years which have not been entirely acceptable to millers and chemists. Are our methods of scrutinizing these varieties unjustifiably severe? I think not, but I am sure that we are subjecting wheats to more numerous and more severe tests than ever before.

Cereal chemists and operative millers are inclined to be critical of any new wheat variety. In fact, it has been my observation that they are prone to expect ideal characteristics in a new variety which do not appear with any degree of uniformity in our most generally approved spring wheat variety, Marquis. It may be well at this point to emphasize the fact that Marquis wheat, although bearing a high reputation, is frequently grown under conditions which result in low flour yield, poor color, high ash, and small

loaf volume. This is a natural effect of environmental conditions and demonstrates the necessity of comparing a new variety with a well-known variety grown under *comparable* conditions. Furthermore, it proves that a single test does not justify generalization.

I have mentioned some of the characters desired by the producer of wheat. I would like to enumerate now milling and baking properties sought by the cereal chemist and his co-worker the flour miller: uniform, plump kernels, to facilitate removal of foreign matter; ease in conditioning and separation of endosperm from offal; high flour yield with production of low ash flour; good color, (creamy, not gray), susceptible to bleaching with any of the common agents; high protein; large loaf volume which is improved by bleaching; quality or strength of gluten which is not impaired by heavy bleaching treatment and will withstand commercial manipulation; gas-producing capacity sufficient to produce uniformly large volume over a long range of fermentation, at the same time giving a loaf with the break, shred and crust color that the baker requires. Add these demands of the commercial mill to those of the grower of wheat and does not the total present difficulties that look almost insurmountable?

To complicate the situation all commercial mills do not have the same viewpoint. This is not unnatural, for mills frequently build up their business by serving a certain class of trade. For example, one mill supplies a housewife trade primarily, another a certain type of commercial bakery which demands a cheap, low protein flour; another serves a trade requiring an exceptionally high protein, strong flour, while still another makes an all-purpose flour in order to draw business from wide sources. The requirements for a mill mix are entirely different in these mills, and consequently their collaboration in testing new wheats in their own laboratories will result in different conclusions, although their several reports might coincide in the details of analysis. I do not offer this as a criticism of such mills. I think it is but a natural outcome of their varied and peculiar requirements for wheat types. It is evidence of the fact that there is a place in the wheat market for wheats of different milling and baking types, just as there is a need for several varieties of wheat to satisfy different agronomic conditions.

I have mentioned standards set up by the cereal chemist. I wish to elaborate this point somewhat before referring to results obtained with a new wheat variety. Yield of flour always will be

the first consideration of the miller, and the value of wheat will be based partly on this property. Naturally the miller desires wheats which will fit into his usual scheme of conditioning. Wheat varieties differ in their capacity for absorbing water during tempering. This should not be viewed too seriously in examining a new wheat, since environmental conditions are liable to cause greater variations in this character than the differences between varieties grown under comparable conditions. Nevertheless this criticism of incompatibility in the mill mix has been raised in the case of Garnet wheat, and the Minnesota Experimental Flour Mill conducted last year an experiment at the request of the Canadian Experimental Farms to compare Garnet and Marquis in conditioning and grinding properties (Sherwood, 1930). In the two lots of wheat tested, Garnet was found to require more water and longer time to bring about the same condition for milling than the Marquis. Mills grinding hard, dry wheat and using a long tempering system would welcome wheat of the type of Garnet, while such wheat would not be acceptable to mills using soft wheat and a short tempering system.

Color of flour is ever important. Fortunately we have a new method for measuring the yellow color (Ferrari and Bailey, 1929) and can now report it in terms of carotin content. New wheat varieties are now being tested for carotin content at the Minnesota Agricultural Experiment Station. Flour is scrutinized for its reaction to various bleaching agents, for the cereal chemist wishes to know whether the color can be removed sufficiently for commercial use without impairing baking properties. This involves bleaching tests with 3 or 4 different agents, followed by numerous baking tests and color determinations. Baking tests by a single method rarely suffice. Resistance to long fermentation is determined by a series of tests in which fermentation time is varied, and frequently a curve is drawn to show the effect of time upon loaf volume. Reaction to oxidizing agents is indicated by measuring the effects of varying increments of potassium bromate. In addition, it is not uncommon to make baking tests at intervals after milling to determine the effects of natural aging. Laboratory tests alone are not relied upon, but doughs of commercial size are carried through the usual commercial manipulation in an effort to detect deficiencies that might have been overlooked in small laboratory doughs.

Comparisons of two varieties are not based upon a single grade of flour. In the Minnesota Experimental Flour Mill it has

been found useful to study the mill streams as well as patent and straight flours in order to secure a complete picture.

I think you will agree that the tests applied by the cereal chemist are more varied, more specific, and more severe than those used to measure the milling and baking properties of wheat 10 or 15 years ago. How are new wheat varieties measuring up to these elaborate standards? I haven't time for a detailed discussion of many varieties. I will mention a few.

Kota wheat, developed in North Dakota, has not become a large factor in the market. It yields well under dry conditions, but lodges badly in many parts of Minnesota, and is very susceptible to smut; hence it is not universally acceptable to farmers.

Quality wheat is not the equal of Marquis in baking properties, and has proved quite variable from different localities, although it mills easily with a large flour yield. Garnet has been found slightly harder than Marquis, but yields large loaves with only fair texture and color. Ceres compares very favorably with Marquis in milling and baking characteristics. It is quite resistant to rust but susceptible to scab and smut.

Reliance, Supreme and Reward are being tested at the several experimental farms. They have shown promise from a milling and baking viewpoint, although not resistant to stem rust. Hope wheat is a new variety developed by McFadden in South Dakota. It is extremely resistant to rust, but susceptible to black chaff. Too little is known of its milling and baking properties, as it has been grown only a short time.

Among the winter wheats Minhardi and Minturki are best adapted to Minnesota conditions because of their winter hardiness. The latter is more widely grown on account of its larger yield per acre. In addition it is resistant to rust and to covered smut. It has good milling and baking properties, compared to other varieties of winter wheat; like all winter wheat grown in the Northwest, however, it is inferior in bread-making properties to the average spring wheat.

I wish to give you a more complete account of Marquillo wheat, a rust resistant variety developed at the Minnesota Experiment Station. Marquíllo is a cross beteen Marquis and Iumillo, originating in 1922. It has proved extremely resistant to stem rust. The average results of several years have shown higher yield per acre than Marquis, and in bad rust years it has exceeded Marquis considerably. Marquillo does not compare favorably with Marquis in southern Minnesota, apparently being much better adapted to

conditions farther North. Over a period of six years Marquillo has exceeded Marquis somewhat in protein content.

Milling and baking tests of this new variety were made in the Minnesota State Testing Mill late in 1929. Two hundred bushels of Marquillo were secured from four different points in the State, and the same quantity of Marquis, for comparison, grown under quite comparable conditions. From two shipping points both varieties had a test weight of about 60 pounds per bushel, while from the other two points both tested about 56 pounds. Consequently two series of tests were made, one with heavy weight, the other with light weight wheat. In all tests Marquillo gave the larger yield of flour. Table I shows the yields.

TABLE I

RESULTS OF MILLING TESTS

	Weight per bushel	Protein (N x 5.7)	Yield		Ash Content	
			Straight Flour	Total Feed	Straight Flour	Patent Flour
	lbs.	%	%	%	%	%
Marquis						
Test 1	55.0		68.7	31.3	.48	.40
Test 2	60.0		71.8	29.1	.45	.37
Average	57.5	13.0	70.2	30.2	.465	.385
Marquillo						
Test 1	55.5		74.4	25.0	.54	.48
Test 2	60.0		76.1	23.2	.48	.43
Average	57.5	14.4	75.2	24.1	.510	.455

Marquillo wheat and flour showed higher ash content, and the flour was more yellow than Marquis in unbleached samples, both straight grade and patent.

Bleaching agents were capable of removing a large portion of the Marquillo color without injuring baking properties, but when the same bleaching treatment was applied to comparable grades of the two varieties Marquis always gave a superior color score. Table II shows the effects of certain bleaching treatments.

TABLE II

CAROTIN CONTENT IN PARTS PER MILLION OF UNBLEACHED AND BLEACHED FLOURS

	Unbleached	Bleached (Dosage per barrel)		
		¾ oz. Beta-Chlora	¾ oz. Beta-Chlora 14 gm. Novadel	2 gm. Agene 14 gm. Novadel
Marquis straight	1.60	1.04	0.42	
Marquillo straight	2.42	1.48	0.48	
Marquis Patent	1.48			0.29
Marquillo Patent	2.23			0.48

Color in these comparative tests was such an important item that instead of relying on two grades of flour the carotin content of the mill streams was determined. The average carotin content of six of the middlings flour streams was: Marquis 1.62, and Marquillo 2.25 parts per million. Marquillo was 40% higher in carotin content.

From the baking standpoint (except color) the Marquillo in these tests appeared acceptable commercially. Absorption was invariably higher than the Marquis, averaging 1.5% higher; volume was invariably higher, averaging 240 cc. Color was 3.0 points lower in unbleached and 2.0 lower in bleached samples; texture was 2.8 points lower unbleached and 1.4 lower bleached. Table III shows comparisons of baking tests.

TABLE III

BAKING TESTS OF MARQUIS AND MARQUILLO FLOURS

	Loaf Volume Patent	Loaf Volume Straight	Abs.	Color		Texture	
				Unbl.	Bl.	Unbl.	Bl.
Marquillo as % of Marquis	114	112	102	97	98	97	98.5

Marquillo was superior in stability as measured by dough action and loaf volume with extended fermentation, and also superior in oven expansion.

These conclusions were confirmed in general by collaborative tests with several commercial mills, where several types of baking tests were used, including 50 pound commercial doughs. Acknowledgment is made to Pillsbury Flour Mills, International Milling Co., Commander-Larabee Milling Co., King Midas Milling Co., Washburn-Crosby Co., Bay State Milling Co., Russell-Miller Milling Co., A. D. Wilhoit Laboratories and Chas. W. Ingman Laboratories.

Marquillo is not the the only variety which warrants tests of such a nature. At University Farm, Minnesota, and at North Dakota Agricultural College there are several promising crosses which appear from small scale tests to have even better baking properties than Marquillo. As soon as sufficient quantities of wheat are available these varieties should be tested thoroughly on a large scale in order that wheat producers, grain dealers and flour millers will have full information concerning their milling and baking properties. It is apparent that plant breeders welcome co-operation in making such tests. There is an opportunity for the technical staffs in the flour and grain industries to assist more than they have in the past, and it is for their best interests to co-operate fully with the experimental agencies developing new wheat varieties.

Literature Cited

Ferrari, C. G., and Bailey, C. H.
1929 The determination of carotin in flour. Cer. Chem. **6**: 347-371.

Hayes, H. K.
1929 The breeding of improved varieties of spring wheat. Cer. Chem. **6**: 483-493.

Sherwood, R. C.
1930 Milling tests of Garnet and No. 2 Northern (Canadian grade) wheat. (Sub-report No. XX, pp. 89-92 in report of L. H. Newman: Overseas tests of the milling and baking qualities of Garnet wheat). Bull. 134, Department of Agriculture. Ottawa, Canada.

Annual Report of the Secretary-Treasurer

M. D. Mize

January 1, 1931

Heretofore, the Secretary-Treasurer's report has always been made at the Annual Convention and all reports drawn up for the fiscal year which extended from one Convention to the next. Sometimes this period was eleven months in length and again as long as thirteen months. This system had the advantage of having the books always thoroughly checked up and audited during the Convention and in case of a change of Secretary-Treasurer with the election of officers, the office could be transfered without delay. However, this system had the disadvantage of giving very little information on the operating revenue and expense during the year or on the net assets of the Association.

Our rapid growth since 1924 has at last caused these disadvantages to far outweigh the advantages. You must remember that our membership as reported herein lacks only thirty-five of doubling our 1924 membership. During the past December, the Executive Committee voted in favor of having the Secretary-Treasurer's Report changed to cover the calendar year. Hereafter, the annual report will appear in the March issue of Cereal Chemistry since about two months are required to procure all outstanding invoices and make the necessary verifications with the records kept by the Cereal Chemistry office.

You have, no doubt, noticed from year to year that the annual report has always assumed a slightly different form. These changes have been necessitated by the rapid growth of the Association as well as by the fact that the original financial reports were merely lists of receipts and expenditures. Such reports were quite sufficient while the membership was in the neighborhood of one hundred members. This year you will find many changes introduced. Many of these have been necessitated by the adoption of the calendar year; while others have been made, after considerable study to determine the future requirements of the report as well as to furnish the maximum amount of information. It is therefore hoped that the form appearing herein may be retained for sometime to come without changes. Only in this way can our membership become thoroughly familiar with the financial activities of the Association.

DETAILED MEMBERSHIP STATEMENT DECEMBER 31ST, 1930

	Total	Active	Corp.	Hon.
Membership, May 1, 1930	410	368	40	2
New members added	21	18	3	. .
Members reinstated	8	7	1	. .
Paid memberships 12-31-30	439	393	44	2

FINANCIAL STATEMENT

December 1, 1929, to December 31, 1930

RECEIPTS 1930				
Cereal Chemistry				
Membership dues.				
Active	$1,725.50			
1931 Active, paid in advance	*350.00*			
Net 1930		$1,375.50		
Corporation	690.00			
1931 Corporation, paid in advance	*250.00*			
Net 1930		440.00		
Subscriptions	1,473.67			
1930 subscriptions received in 1929	375.50			
1931 subscriptions paid in advance	*495.00*			
Net 1930		1,354.17		
Reprints, back numbers, etc.	698.97			
1930 Accounts Receivable	165.81			
1929 Reprints, etc., paid in 1930	*71.55*			
Net 1930		793.23		
Advertising	1,177.50			
1930 Accounts Receivable	140.50			
1929 Advertising paid for in 1930	*192.00*			
Net 1930		1,126.00		
Interest on Invested Funds		213.75		
Transferred from Association Funds		300.00		
Miscellaneous Income		11.21		
Total Net Receipts 1930			$5,613.86	
Association				
Membership dues (Active)	1,724.68			
1931 Active Membership dues paid in advance	*350.00*			
Net 1930		1,374.68		
Application Fees	113.75			
1931 Application Fees paid in advance	*3.00*			
Net 1930		110.75		
Interest on Invested Funds		100.67		
Total Net Receipts 1930			1,586.10	
Book of Methods Reserve Fund			233.08	
Experimental Baking Fellowship Fund			1,045.91	
Loss 1930				$454.09
TOTAL RECEIPTS OF ALL ACCOUNTS 1930			$8,478.95	
TOTAL LOSS OF ALL ACCOUNTS				$454.09

DISBURSEMENTS 1930

Cereal Chemistry

Cost of editing, printing and mailing Cereal Chemistry and reprints	$6,050.38		
1930 Accounts Payable	18.00		
1929 Editing expenses paid in 1930	*393.57*		
1929 Reprint expenses paid in 1930	*69.35*		
1929 Commission paid in 1930 on advertising	*48.60*		
Total Net Disbursements 1930		$5,556.86	
Profit in 1930			$57.00

Association

Expenses of President's, Vice-President's Office; News Letters	291.98		
Expenses of Office of Sec'y-Treas	380.07		
Committee expenditures	197.94		
Convention expenses	124.35		
Transferred to Cereal Chemistry	300.00		
Total Net Expenditures 1930		1,294.34	
Profit in 1930			291.76
Book of Methods mailing		15.00	
Profit in 1930			218.08
Experimental Baking Fellowship Fund; Sept. 1, 1930, to Feb. 28, 1931		1,500.00	
TOTAL DISBURSEMENTS OF ALL ACCOUNTS		$8,366.20	
TOTAL PROFIT OF ALL ACCOUNTS			$566.84
NET PROFIT		$112.75	$112.75

PROFIT AND LOSS ACCOUNT

Cereal Chemistry Assets 1929	$2,559.52	
Profit 1930	57.00	
Cereal Chemistry Assets 1930		$2,616.52
Association Assets 1929	1,034.12	
Profit 1930	291.76	
Association Assets 1930		1,325.88
Book of Methods Reserve Fund 1929	111.61	
Profit 1930	218.08	
Book of Methods Reserve Fund 1930		329.69
Convention Reserve Fund 1929	452.78	
Convention Reserve Fund 1930		452.78
Experimental Laboratory Baking Fund 1929	3,260.15	
Loss 1930	454.09	
Experimental Laboratory Baking Fund 1930		2,806.06
Total Assets December 31, 1930		$7,530.93

Note: All amounts in italics are negative amounts and are subtracted from the other amounts in the same column.

In the annual report of Secretary-Treasurer, May 7, 1930, Assets of Cereal Chemistry were shown as.. $3,182.99 in December, 1929;
but this figure included..... 375.50 of 1930 income received
in advance which left..... 2,807.49 ; further this figure did not
include 1929 expenses of..... 511.52 which were not paid until 1930;
thus the adjusted balance should
have been ..... 2,295.97 ; further still, this figure
did not include..... 263.55 of 1929 income that was not
paid until 1930. Therefore..... 2,559.52 was the correct amount of Assets

ASSETS AS OF DECEMBER 31ST, 1930

U. S. National Bank—Savings Dept.	$586.34
First National Bank—Savings Dept.	2,544.24
U. S. National—Checking Account	48.91
Inter City National Bank in Kansas City, Missouri	411.13
Petty Cash Fund in Minneapolis	100.00
Building & Loan Stock in Kansas City	2,000.00
Building & Loan Stock in Omaha	1,500.00
Building & Loan Stock in Minneapolis	1,500.00
1930 Accounts Receivable	306.31
GROSS ASSETS	$8,996.93
LIABILITIES.	
1931 Income received in 1930	1,448.00
1930 Accounts Payable	18.00
NET ASSETS	$7,530.93

ERRATA

Biometric Analysis of Cereal-Chemical Data I. Variation. Alan E. Treloar. Cereal Chemistry, Vol. VIII, January, 1931.

p. 82—4th line from bottom, $P.E._x$ should read $P.E._{\bar{x}}$.

p. 86—Transpose last four lines to position immediately below Figure 4 on same page.

Contribution to the Colloid Chemistry of Gluten III. H. L. Bungenberg de Jong and W. J. Klaar. Cereal Chemistry, Vol. VII, November, 1930.

p. 589—In Fig. 1 the solid line represents the influence of *acetone* concentration and the dotted line the influence of *alcohol* concentration.